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THE BOTANICAL GAZETTE

EDITOR: JOHN M. COULTER

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EDITED BY

JOHN M. COULTER

With the assistance of other members of the botanical staff of the University of Chicago

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THE
BOTANICAL GAZETTE

March 1923

EFFECT OF ANDROPOGON SORGHUM ON SUCCEEDING CROPS OF TRITICUM SATIVUM VULGARE

CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY 301

M. C. SEWELL

(WITH TWELVE FIGURES)

Since the introduction of sorghum into the Great Plains area, it has commonly been observed that *Triticum sativum vulgare* planted in the fall after *Andropogon sorghum* does not yield as well as when planted after *Zea mays*. Reports of this effect of kafir on wheat have been made in various state experiment station bulletins, notably by TEN EYCK and SHOESMITH (**20**), BENNETT (**1**), SNYDER and OSBORN (**19**), and VINALL and GETTY (**21**). There is an extensive literature on the subject of soil fertility and the influence of one crop upon another, but only such papers as have a direct bearing upon the work will be cited. The literature suggests five possible causes of the harmfulness of kafir: depletion of minerals, toxic root excretions, toxic products of decay, diseases associated with the crop, and the effect of soil protozoa and micro-organisms.

HOLTER and FIELDS (**8**) found that kafir has a much higher ash content than corn. In a series of analyses made at the Oklahoma Agricultural Experiment Station and published in the Twenty-ninth Annual Report, the ash analyses of kafir show that this crop removes a great deal of potassium and phosphorus out of the soil. The water soluble nitrogen was found to be very large in the young

plant, but to decrease very rapidly as the plant nears maturity. WILLAMAN and associates (23) decided that the plant absorbs practically all of its mineral requirements, including nitrogen, during the early stages of growth, that it lays down the necessary structures of protein and fiber during these stages, and that during the final maturation periods all the energies of the plant are directed toward developing the seed. These facts would be contrary to the prevalent idea that kafir is extracting elements of nutrition late in the fall as the crop matures. While these references indicate that the kafir plant grown as a crop may extract more of the mineral elements of nutrition from the soil than corn, it is difficult to conceive of one crop of kafir exhausting the supply to the detriment of the succeeding crop of wheat.

A number of investigators have developed the idea that plants might affect succeeding crops by toxic root excretions. FLETCHER (4), working at Surat, India, reports that a row of *Sesamum indicum* when grown at a distance of two feet from a row of sorghum (great millet) will not mature, the plants dying after reaching a height of a few centimeters. *S. indicum* was used because of its extreme sensitiveness.

WHITNEY and CAMERON (22) postulated the theory that the cause of low production in soils is frequently due to the fact that plants in their decomposition products give off organic substances that are toxic to the growth of plants, and that practically all soils contain sufficient plant food for crop yields. Co-workers of WHITNEY and CAMERON, LIVINGSTON, BRITTON, and REID (10), studied the properties of an unproductive soil, a silt loam from Takoma Park, Maryland. An organic substance deleterious to plants was found in this soil. SCHREINER and REED (18) present evidence that the diminished yields of a second crop are not due to the removal of plant nutrients by the first crop. SCHREINER and SHOREY (16) considered the isolation of a definite organic compound, dihydroxystearic acid, injurious to plant growth, as tangible proof of the injurious organic compounds, and that their work laid the foundation for the rational study of unproductive, exhausted soils. Additional conclusions of WHITNEY and his colleagues were that a soil deteriorates in fertility and ceases to yield normal crops, not

because of any lack of plant food brought about by the continuous withdrawal of the original stock in the soil, but because of the accumulation of injurious substances excreted by the plant itself. These toxins are held to be specific to each plant, but gradually removed from the soil by processes of decay.

With extensive study and experimentation, PICKERING (11) found that the growth of one plant may have a deleterious effect on another plant grown in close proximity. In experiments on the effect of grass on trees (12), it was concluded that this effect could not be attributed to simple causes such as root competition, increased evaporation, differences in temperature, or aeration of the soil, but must be due to some toxic action exerted by the grass. The toxic matter formed by grass was quickly oxidizable. Toxic grass leachings when exposed to the air and thus oxidized had a beneficial effect upon tree growth. PICKERING considered that there was no reason for assuming the excretion of any toxic matter from a plant, but that the débris from the growing plants is probably sufficient to account for the formation of the toxin.

One of the main reasons given for crop rotation is that thereby a crop with certain specific characteristics is followed by another with quite different requirements. The Rhode Island Experiment Station inaugurated a long time field experiment in order to gain information upon the principles of rotation. The results are reported by HARTWELL and DAMON (6), also by HARTWELL, PEMBER, and MERKLE (7). The field experiments consisted of growing each of sixteen crops two out of three years on the same soil, and every third year planting one of those crops uniformly over the entire area. The amount of nutrients removed by the crops was determined, but it was not found that the crops which removed the largest amount of the scarcest nutrients were the ones which had the most depressing effect on a succeeding crop. This means that the smallest yields may not occur after the crops which removed the largest amount of even the most needed nutrient.

As evidence that diseases in the soil may be a factor causing unproductiveness, there are the cases of flax wilt, wheat blight, and tobacco root rot. BOLLEY (2) determined that flax wilt is caused by a parasitic fungus which can exist for years in the soil. Con-

cerning the problem of wheat deterioration, BOLLEY (3) concluded that constant wheat cropping tends to introduce with the seed, stubble, and roots, a number of wheat disease-producing fungi. These destroy, blight, or rot off the roots of wheat plants and live internally in the straw and the seeds. JOHNSON and HARTMAN (9) found that continuous cropping of tobacco results in low productivity because of a parasite which infests the soil and produces the *Thielavia* root rot of tobacco. The result is a stunting of the plant due to a reduced root system.

Relative to the action of soil protozoa and micro-organisms on the productivity of soils, HALL (5) states that if we assume that the persistence in the soil of obscure diseases appropriate to the particular plant can be neglected as the cause of low productivity, there still remains some unexplained factor arising from a plant's growth which is injurious to a succeeding crop, and that this may be either the toxin of WHITNEY'S theory or some secondary effects due to the competition of injurious products of the bacteria and other microflora accumulating in the particular soil layer in which the roots of the crop chiefly reside.

RUSSELL (14) and other workers have shown how greatly a disturbance of the normal equilibrium of the flora of the soil may affect its fertility. Partial sterilization, such as is brought about by heating the soil to 98° for ten hours, will double the yield of the succeeding crop, and will show a beneficial effect upon the fourth crop after heating. Exposure to vapors of volatile antiseptics like toluene or carbon bisulphide, which are afterward entirely removed by exposure, will increase the yield in a similar but smaller degree. Even drying the soil appears to have an influence upon its fertility. RUSSELL and PETHERBRIDGE (15) studied sickness in certain glass house soils, and concluded that it is conditioned by an accumulation of insect and fungoid pests and by a lowered bacterial efficiency. The lowering of the bacterial efficiency was considered due to the accumulation of a factor detrimental to bacteria. This factor is capable of growth when introduced into partially sterilized soils. In all respects its properties agree with those of a protozoan. They found soil sickness in tomato and cucumber houses to be effectually treated by sterilization.

The purpose of this investigation was to determine the effect of kafir crops on following wheat crops, and whether the generally experienced deleterious effect is due to a depletion of water and nutritive elements in the soil, or to the formation of toxic organic substances in the decomposition products of the crop residue.

Method

Field plots were located of one-twentieth acre in size, two of which were cropped each year to corn and two to kafir, and all alternated with winter wheat. Measurements were made of the comparative yields of corn and kafir and of the yields of the succeeding wheat crops. Soil samples were taken to a depth of three feet during the summer period, and in the fall before seeding wheat, to determine the amount of nitrate nitrogen. The latter was used as an index of the available nutrient materials. In taking soil samples, a composite was made of three cores on each plot for each section of soil sampled. A steel tube was used to extract the cores. For the moisture determination, the cores were passed through a coarse sieve, the soil rolled in an oil cloth, and 50 gm. duplicate samples weighed out in aluminum dishes and placed in an electric oven maintained at 108°–110° C. For the nitrate determination, duplicate 100 gm. samples of the sieved cores were weighed into glass stirring vessels. A small amount of calcium carbonate, to facilitate the flocculation of the soil, and 500 cc. of distilled water were added to each vessel. The samples were stirred for three minutes with a mechanical stirring machine, and then filtered through Pasteur Chamberlain filters. From the filtrates nitrates were determined by the phenoldisulphonic acid method, using a DeBosq colorimeter.

Greenhouse studies conducted during the fall and winter consisted in part of growing wheat seedlings in water cultures after the method of SCHREINER and SKINNER (17). Soil solutions from the corn and kafir field plots were used for the water cultures, and various nutrient salt solutions were added to them. The soil solutions were obtained by leaching, filtering through porcelain, and by centrifuging. Separate series of duplicate cultures were conducted with each of these methods of obtaining a solution. Pot cultures were set up, using soil and stubs of corn and kafir and grow-

ing wheat with various fertilizer treatments. In one instance oats were grown in place of wheat. Pot cultures were also employed with leaching trays as described by PICKERING. Kafir and corn were grown in the trays with and without perforated bottoms, and wheat was subsequently planted in the soil below, which received the percolations from the kafir and corn soil.

The ammonifying and nitrifying power of the soil was determined after it had been cropped respectively to corn and kafir. Samples of soil weighed as the equivalent of 25 gm. of dry soil were incubated in glass bottles, with the following treatments: (1) maintained at moisture condition of the field at the time of sampling; (2) maintained at optimum moisture; (3) maintained at optimum moisture plus 1 gm. of $CaCo_3$; (4) maintained at optimum moisture plus 1 gm. $CaCo_3$ and 25 gm. dried blood. The bottles were lightly stoppered with cotton and incubated for periods of one and of four weeks. The determination for ammonia was made by the aeration method of POTTER and SNYDER (13). As in the case of the field samples, nitrate determinations were made according to the phenol-disuphonic acid method.

Results

FIELD STUDIES

TABLE I

YIELDS OF WINTER WHEAT GROWN AFTER CORN AND KAFIR CROPS, 1916–1921, MANHATTAN, KANSAS

YEAR	CORN SOIL		KAFIR SOIL	
	Grain (bushels per acre)	Straw (pounds per acre)	Grain (bushels per acre)	Straw (pounds per acre)
1916.....	27.8	.....	20.3	.....
1917.....	15.2	1,985	17.3	2,570
1918.....	13.9	2,425	8.2	2,082
1919.....	34.0	2,049	31.1	1,872
1920.....	12.3	.....	10.5	.....
1921.....	5.2	750	3.6	710
Average 6 yrs..	18.1	1,802*	15.1	1,808

* Average of four years.

The comparative yields of corn and kafir are given in table II. These data are based upon the weight of field-cured fodder. In

1921, the kafir and milo fodder was analyzed for the percentage of fertilizing elements. The results of the determinations are shown in table III. Typical data obtained on the bacteriological determinations are incorporated in tables IV and V.

TABLE II

COMPARATIVE YIELDS IN POUNDS PER ACRE OF CORN AND KAFIR

YEAR	CORN SOIL		KAFIR SOIL	
	Grain	Fodder	Grain	Fodder
1916	2,357	7,760	2,552	10,960
1917	2,856	13,380	2,025	22,110
1918*				
1919	1,180	5,450	1,632	10,050
1920	1,908	6,451	2,015	8,904
1921	3,063	7,422	2,922	9,435
Average 5 yrs.	2,272	8,092	2,229	12,292

* Corn cut for silage because of drought; kafir killed by early frost.

TABLE III

PERCENTAGE OF FERTILIZING ELEMENTS IN CORN AND KAFIR FODDER, CROP OF 1922

Fodder	Nitrogen	Phosphorus	Potassium
Kafir	1.42	0.243	1.05
Corn	1.48	0.218	0.77

TABLE IV

AMMONIFICATION IN CORN AND KAFIR SOIL; AVERAGES OF DUPLICATE TREATMENTS; 0.05 GM. N ADDED IN DRIED BLOOD

SOIL	PERCENTAGE OF N AMMONIFIED			
	Soils incubated 1 week		Soils incubated 4 weeks	
	Before cropping	After cropping	Before cropping	After cropping
Corn	5.4	17.9	2.7	17.3
Kafir	5.7	18.7	3.3	17.2

Although moisture and nitrate determinations were made for field samples taken monthly in sections to a depth of three feet, it will suffice to present (in table VI) the data for the surface foot at

TABLE V

NITRIFICATION IN CORN AND KAFIR SOILS; AVERAGES OF DUPLICATE TREATMENTS BEFORE AND AFTER CROPPING

SOIL	NATURAL MOISTURE		OPTIMUM MOISTURE		OPTIMUM MOISTURE $CaCO_3$		OPTIMUM MOISTURE $CaCO_3$, DRIED BLOOD	
	Before	After	Before	After	Before	After	Before	After
Corn	17.1	8.9	20.6	7.0	0*	8.9	131.2	17.3
Kafir	8.2	4.9	13.2	6.0	0*	9.3	123.2	17.2

* No determination.

TABLE VI

SOIL MOISTURE AND NITRATE DETERMINATIONS OF SAMPLES FROM SURFACE FOOT OF SOIL

YEAR	CORN SOIL		KAFIR SOIL	
	Moisture per cent dry weight	Nitrates p.p.m.	Moisture per cent dry weight	Nitrates p.p.m.
1916	25.2	29.6	24.0	28.2
1917	16.8	6.6	15.8	6.4
1918	20.5	67.7	20.0	48.5
1919	18.7	63.6	15.1	38.9
1920	16.7	31.1	14.8	34.6
1921	18.3	3.5	16.7	2.1

TABLE VII

AVERAGE MOISTURE AND NITRATES IN DEPTH SAMPLED AT SEEDING TIME OF WHEAT

DATE	AVERAGE DEPTH (feet)	MOISTURE PER CENT		NITRATES P.P.M.	
		Corn	Kafir	Corn	Kafir
1915	2	16.2	17.8	1.5	1.4
1916	6	18.6	20.3	20.5	24.9
1917	3	18.1	18.2	3.1	3.2
1918	3	17.9	17.9	29.8	23.4
1919	3	17.2	16.3	28.1	18.2
1920	3	16.7	16.0	14.5	9.9
1921	3	26.0	25.1	5.8	5.5
Average	.....	18.7	18.5	13.9	11.3

the time of planting wheat in the fall after the corn and kafir crops, and for the average of the total depth sampled. The average moisture and nitrates in the full depth sampled at seeding time of wheat are given in table VII.

GREENHOUSE STUDIES

In order to determine whether sorghum soils possessed any toxicity to wheat, seedlings were grown in water cultures composed of soil solutions from corn and kafir soils. Nutrients were added to these solutions after the method of SCHREINER and SKINNER (17). Employing 20 per cent variations in concentrations, twenty-one bottles were required for a set of corn and kafir soil solutions. Each treatment was duplicated. For these cultures, 250 cc. wide-mouthed glass bottles were used. Flat corks were notched to hold the sprouted seedlings. The concentration of the nutrient solutions was 80 ppm. The stock nutrient solutions were prepared with $CaH_4(PO_4)_2$, $NaNO_3$, and K_2SO_4. The composition of the various solutions varied by 20 per cent differences in composition. Of the three duplicate series of water cultures conducted, the one in which the soil solutions were obtained by certrifuging is probably the most reliable. The data thus secured are visualized by figures. Circles in equilateral triangles represent the position of the wheat culture in the triangle and thus its fertilizer ratio. The diameter of the circle represents the weight of the plants produced by the respective nutrient solutions. The wheat seedlings were grown for a period of thirty days. The results are presented in figs. 1–6. The weights of wheat seedlings grown in soil solutions of corn and kafir soils without the addition of nutrients are given in table VIII.

Table IX gives the total weights of plants produced in the twenty-one cultures of each triangle. Chemical analyses were made of the various soil solutions. Table X gives the results of determinations obtained with the centrifuged solutions.

Soil from the corn and kafir plots was brought into the greenhouse in the fall (November), passed through a 2 mm. sieve, and placed in twelve stone jars fitted with proper drainage. At the time of obtaining this soil, the moisture content was 12.8 per cent for the corn soil and 10.8 per cent for the kafir soil. The jars

contained an average of 15.7 kg. of soil, and were maintained at an optimum moisture content of 27 per cent. The six treatments for corn and kafir soil were: no. 1, complete fertilizer of N–P–K;

TABLE VIII

WEIGHTS OF WHEAT SEEDLINGS GROWN IN SOLUTIONS OF CORN AND KAFIR SOILS

TREATMENT	WEIGHT OF TOPS (GM.)				WEIGHT OF ROOTS (GM.)	
	Corn soil		Kafir soil		Corn soil	Kafir soil
	Green	Dry	Green	Dry		
Leachings	2.18	0.33	2.05	0.31	0.23	0.25
Filtered	1.83	0.35	1.68	0.34	0.23	0.20
Centrifuged	1.51	0.30	1.47	0.29	0.27	0.24

TABLE IX

TREATMENT	WEIGHT OF TOPS (GM.)				WEIGHT OF ROOTS (GM.)	
	Corn soil		Kafir soil		Corn soil	Kafir soil
	Green	Dry	Green	Dry		
Leachings	50.64	7.14	44.48	7.37	5.24	5.27
Filtered	78.01	10.37	80.54	11.26	6.40	6.66
Centrifuged	83.19	12.51	79.65	12.69	7.65	6.86

TABLE X

CHEMICAL ANALYSES OF SOLUTIONS OBTAINED BY CENTRIFUGING; RESULTS EXPRESSED IN PARTS PER MILLION (1000 C.C. OF SOLUTION EQUIVALENT TO 400 GM. SOIL)

Soil solution	Colloidal material	Total soluble salts	Nitrates	Phosphorus	Silica	Potassium	Calcium
Corn	921.7	671.5	2.8	0.92	160.0	9.2	3.1
Kafir	1088.8	677.2	2.1	0.90	120.4	14.2	3.5

no. 2, phosphorus; no. 3, nitrogen; no. 4, potash; no. 5, lime; no. 6, no treatment. Where these fertilizers were added, the growth of oats in the kafir soil exceeded the growth in the corn soil. This was also the case with lime applications.

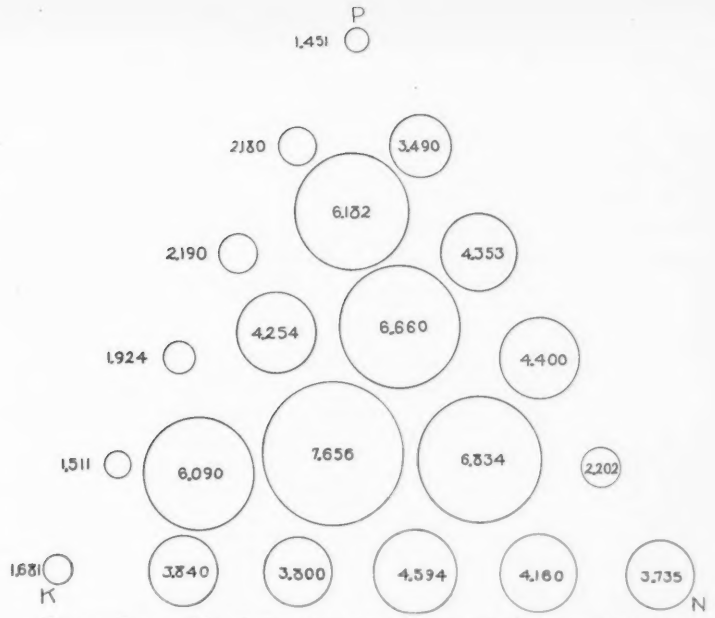

FIG. 1.—Green weight of wheat tops grown in corn soil solution with additions of nutrients.

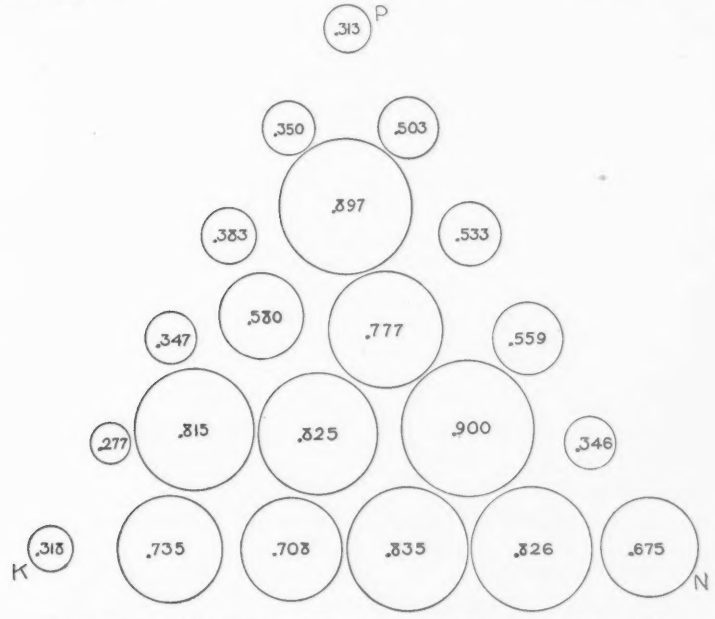

FIG. 2.—Dry weight of wheat tops grown in corn soil solution with additions of nutrients.

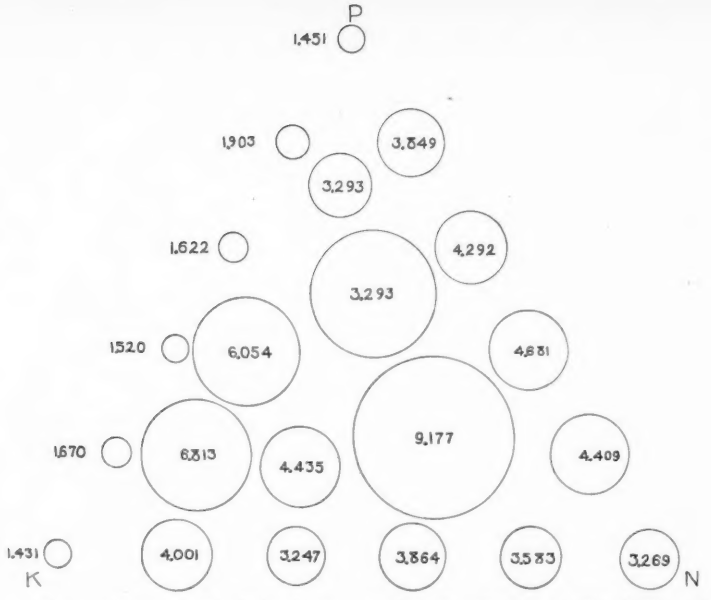

FIG. 3.—Green weight of wheat tops grown in kafir soil solution with additions of nutrients.

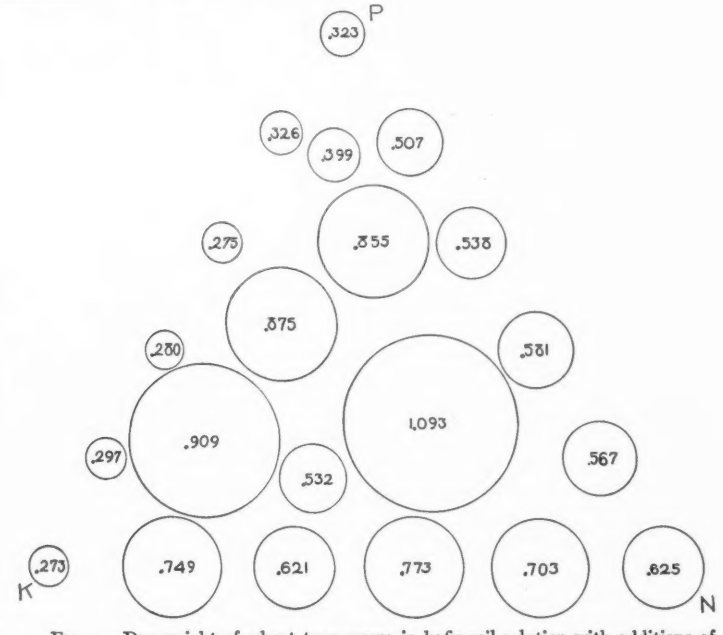

FIG. 4.—Dry weight of wheat tops grown in kafir soil solution with additions of nutrients.

Several years later four large containers, each holding about 350 lb. of soil, were placed in the greenhouse. Two were filled with corn soil and two with kafir soil, and wheat was planted. After the first sixty days there was considerable difference in growth, as recorded in fig. 5, the wheat in the corn soil being much the better. The weight of wheat produced is presented in table XI.

TABLE XI

WHEAT IN LARGE CONTAINERS

Treatment	Total weight of wheat at harvest (gm.)	Grain weight (gm.)
Kafir soil	154 } Mean 132	23
Kafir soil	110 }	18
Corn soil	325 } Mean 361	62
Corn soil	397 }	60

The difference in mean yields, total weight, is 129. The probable error of the difference is 35. A significant difference should be 3.8 times the probable error. Since 129 equals 3.7 times the probable error of 35, we have a significant variation in yield. Other

TABLE XII

POT CULTURES WITH WINTER WHEAT

Treatment	Total weight of ripe wheat (gm.)
Corn soil	35 } Mean=40.3 ±4.37
Corn soil	43 }
Corn soil	43 }
Kafir soil	17 } Mean=22.6 ±6.92
Kafir soil	11 }
Kafir soil	40 }
Corn soil+kafir roots	32 } Mean=33.6 ±3.76
Corn soil+kafir roots	26 }
Corn soil+kafir roots	43 }

pot cultures were conducted in three gallon jars growing wheat, as explained in table XII.

Applying PETER'S formula of probable error, the difference in means between corn and kafir soils is 16.7. The probable error of the difference is 8.6. The difference should be 3.8 times the probable error to be significant.

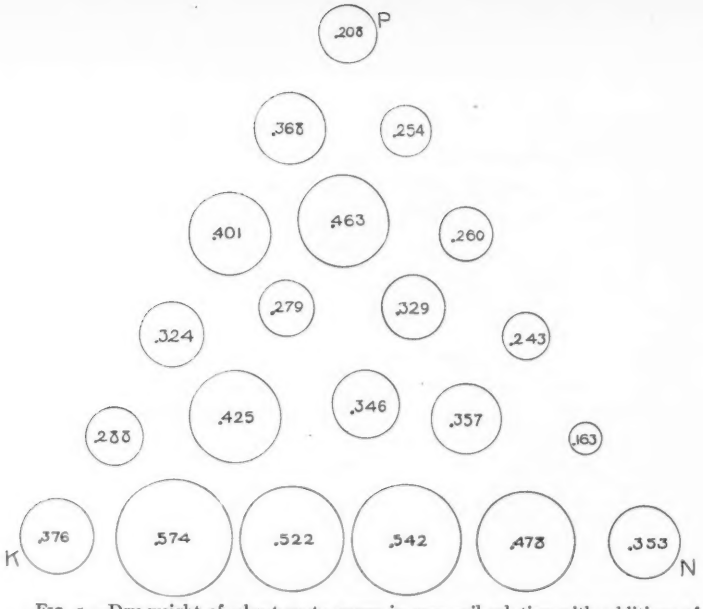

FIG. 5.—Dry weight of wheat roots grown in corn soil solution with additions of nutrients.

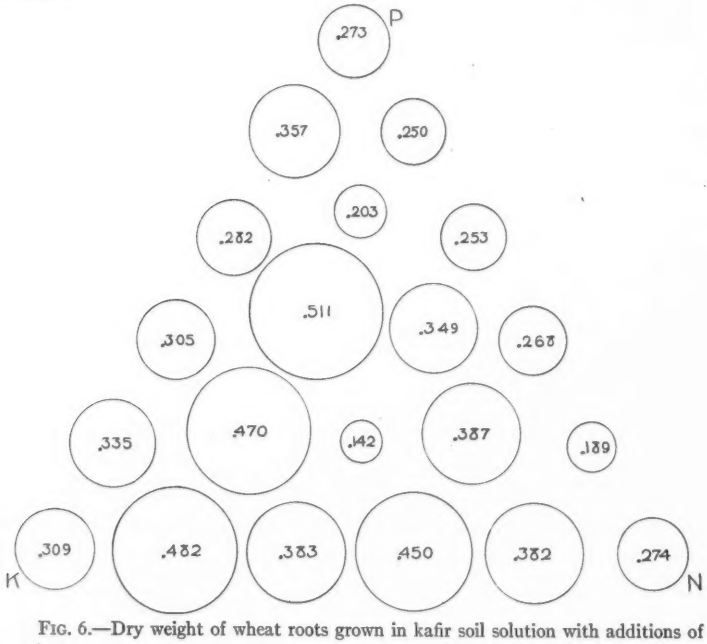

FIG. 6.—Dry weight of wheat roots grown in kafir soil solution with additions of nutrients.

Another phase of the greenhouse work was the employment of pot cultures, using the type of percolating trays used by PICKERING (12) at the Woburn (England) Experimental Fruit Farm. The trays, which were made of galvanized tin, were 10 inches in diammeter and 5 inches in depth, with an opening through the center

TABLE XIII

POT CULTURES WITH PICKERING TRAYS

Weight of plants at maturity	Mean weight	Weight of grain	Mean weight of grain
	Kafir in perforated tray		
24.5	.....	3.0	.....
28.0	.....	3.5	.....
18.0	23.9	2.5	2.8
28.0	.....	3.0	p.e.=0.185
21.0	.....	2.0	.....
	Kafir in solid bottom tray		
20.0	.....	1.5	.....
24.0	.....	3.0	.....
28.0	24.2	4.0	3.5
31.0	.....	5.5	p.e.=0.422
18.2	.....	3.5	.....
	Corn in perforated tray		
24.0	.....	4.0	.....
22.8	.....	4.0	.....
17.6	21.8	3.0	3.7
23.0	.....	3.5	p.e.=0.152
22.0	.....	4.0	.....
	Corn in solid bottom tray		
22.4	.....	3.0	.....
25.0	.....	5.0	.....
20.0	23.8	3.0	3.6
17.0	.....	2.5	p.e.=0.388
35.0	.....	4.5	.....

3.5 inches in diameter. They were placed inside of ordinary 3 gallon stoneware jars filled with soil to a height that brought the top of the trays even with the top of the jars. The trays contained soil in which corn and kafir respectively were grown. Wheat was planted in the soil contained in the stone jar, exposed through the hole part of the tray. The results are given in table XIII.

Comparing corn and kafir in perforated trays, the probable error of the difference is 0.212. The difference in mean yields of grain is 0.9, which is 4.24 times the probable error of the difference, and hence significant. Comparing corn and kafir in solid bottom trays, the probable error of the difference is 0.574 for the grain weights, and the difference in means is 0.1. (To be significant, this difference should be 2.182.)

Discussion

The yield of winter wheat grown after kafir has been three bushels less than when grown after corn, considering the average measured yields for a period of six years. During this period, kafir produced 4200 lb. more of field cured dry matter than corn on the acre basis. From the fodder analyses stated in table III, calculations have been made of the fertilizer constituents removed by the average yields of corn and kafir and were found to be as follows:

	Nitrogen (lb.)	Phosphorus (lb.)	Potassium (lb.)
Kafir	174.5	29.8	129.0
Corn	119.7	17.6	62.3
Difference	54.8	12.2	66.7

To supply this difference in nutritive elements removed, it would take 172 pounds of sodium nitrate, 76 pounds of 16 per cent acid phosphate, and about 160 pounds of potassium sulphate. It would thus seem that in producing more dry matter kafir does remove more fertilizing elements than corn. Liebig maintained that the proper fertilizer for any particular crop must contain the amounts of nitrogen, phosphoric acid, potash, and other constituents which are withdrawn from the soil by a typically good yield of the plant in question, but to show that the amounts of nutritive elements required are not indicated by the composition of the crop, Hall (5) cites the yields, fertilizing experience, and composition of wheat and barley, at the Rothamstead Station. The results of field experiments, confirmed by ordinary farming experience, show that the yield of wheat is chiefly determined by the supply of nitrogen; phosphoric acid is of secondary importance, and only exceptional

soils will give any return for the application of potash. The yield per acre and the composition of the wheat and barley crops are as follows:

	Yield of grain (bushels per acre)	Lb. per acre removed		
		N	P_2O_5	K_2O
Wheat	36	50	21	29
Barley	48	49	21	36

With barley, although its composition is similar to that of wheat, the results are very different. Nitrogen is still the important element in nutrition, but phosphoric acid has equally marked effects, although potash counts for little or nothing. The analysis of the barley plant would indicate that it requires nitrogen in the largest amounts, then potash, and least of all phosphoric acid. The phosphoric acid requirement of barley is probably due to its being a shallow rooted crop.

The Swede turnip crop is given as another noteworthy example. An analysis of a representative yield would show it to withdraw from the soil 150 pounds per acre of nitrogen, 30 pounds of phosphoric acid, and 120 pounds of potash, yet the ordinary fertilizer for the Swede crop will consist of only a small amount of nitrogen, rarely or never potash, but a considerable quantity of phosphoric acid.

In the light of this Rothamstead experience, the fact that kafir removes more nutritive elements than corn can be discountenanced as the explanation of the effect of kafir in depressing the wheat yield. The bacteriological determinations do not signify that the ammonifying and nitrifying power of the soil has in any way been impaired by the growth of kafir as compared with corn (tables IV and V).

The moisture data for the surface foot of soil in table VI shows a slight difference favoring the corn soil. A difference of two per cent moisture in the surface soil is probably sufficient to effect the early growth of wheat at planting time. When the total depth of three feet of soil is considered, the amount of moisture in the kafir soil has been as much as in the corn soil (table VII).

In table VI, the nitrates in the surface foot are considerably less in 1918 and 1919 in kafir soil, but other years vary but little from the corn soil. The average nitrates in a three foot depth of soil are also less in the kafir soil these same years. Fig. 7 compares

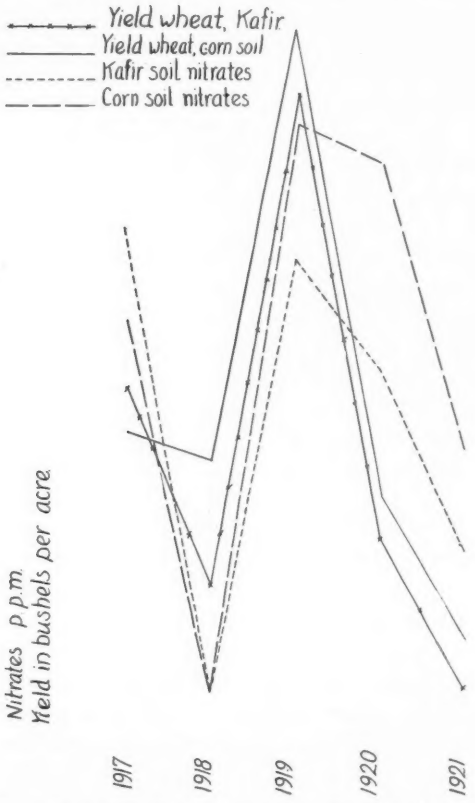

FIG. 7.—Comparison of relation of nitrates at seeding time to yield of wheat

the relation of nitrates in the total depth of soil sampled at seeding time with the yield of wheat the year following. In 1917, the nitrates ran a little greater in the kafir soil, and the yield of wheat was larger that year after kafir than after corn. In 1918, with the nitrates the same in amount, there was a wide variation in yield.

In 1920, although the yields varied but little, there was a wide difference in the amounts of nitrates. In 1921, the nitrates were considerably higher than in 1918, but the yields were much less than in 1918. The fall in the yield curves from 1919 to 1920 is much greater in rate than the drop in the nitrate curves. Although there

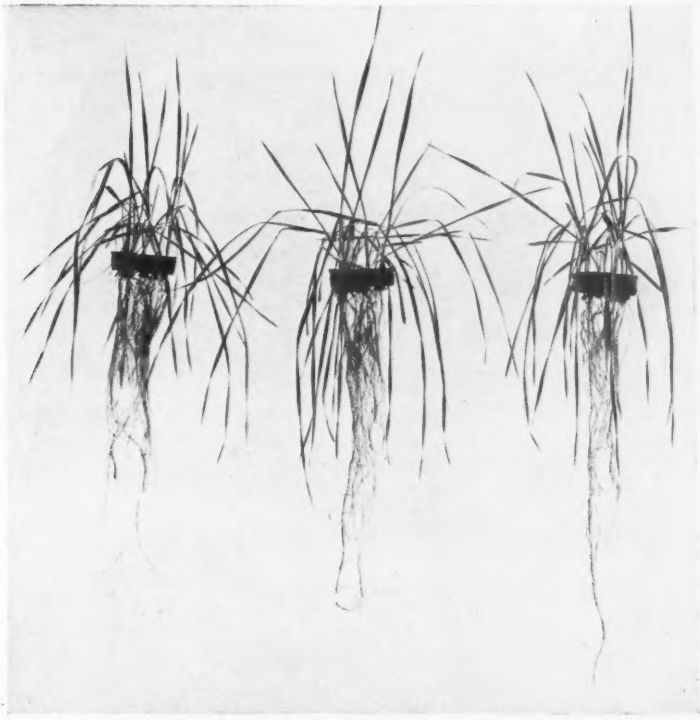

FIG. 8.—Wheat seedlings grown in corn, kafir, and milo soil solutions

is evidence of a general correlation between the yield curves and nitrate curves, it seems doubtful that the nitrates have been the determining factor in the yield variations. Fig. 8, showing the wheat seedlings grown in corn, kafir, and milo soil solutions, does not give any evidence of toxic properties of the sorghum soil solutions. Figs. 1–6, representing the growth of seedlings in triangles

with various combinations of the fertilizing elements, do not show any great variation in the effect of these elements in kafir soil solution compared with corn soil solution. After studying the work of PICKERING, it would seem that the method used in obtaining soil solutions afforded such an opportunity for oxidation that a toxic effect could not be expected even though present in the field soil. PICKERING found that leachings from growing grass, percolating directly into the soil, inhibited the growth of orchard trees, yet when these leachings were collected, aerated, and then applied to the trees, growth was promoted.

In uncompleted work, leachings from kafir soil with its residue have been secured by means of glass tubes attached to the bottoms of elevated jars. With such a system the leachings had but little chance for oxidation. Kafir soil leachings obtained in this manner produced a stunted root development on wheat seedlings, and caused a reddish brown precipitate in the solution. When the kafir soil solutions were aerated by drawing a current of air through them, the stunting effect was not so marked. Fig. 12 illustrates the results obtained.

Concerning the extensive water cultures experiments conducted and reported here, however, it can only be said that with this method kafir soil did not give evidence of toxicity. The solutions in the figures cited were obtained by centrifuging soil solutions at the rate of 3000 revolutions per minute. Fresh soil samples were brought in from the field plots every three days, and the solutions were then changed in the cultures. From composite soil samples, 100 gm. of soil were taken and stirred with 500 cc. of water, and then centrifuged. In tables VIII and IX there is a summary of the growth of the wheat seedlings in the three methods of obtaining the soil solutions in the three different series of experiments conducted. In each instance the kafir soil solutions produced practically the same total weight of seedlings as the corn soil solutions.

The analyses made of the centrifuged solutions do not indicate any difference between the corn and kafir soil solutions except in the matter of potash and silica, these being less in the kafir solutions. Where fertilizing elements were applied to pot cultures of corn and kafir soils, however, potash on kafir soil did not give much increase

in the growth of oats. Lime alone gave an increase, although the soils are not acid in reaction. Nitrogen and phosphorus applied to the oats grown in kafir soil produced a growth excelling the untreated corn soil, but the analyses of the soil solutions did not show that kafir soil contained less nitrogen and phosphorus.

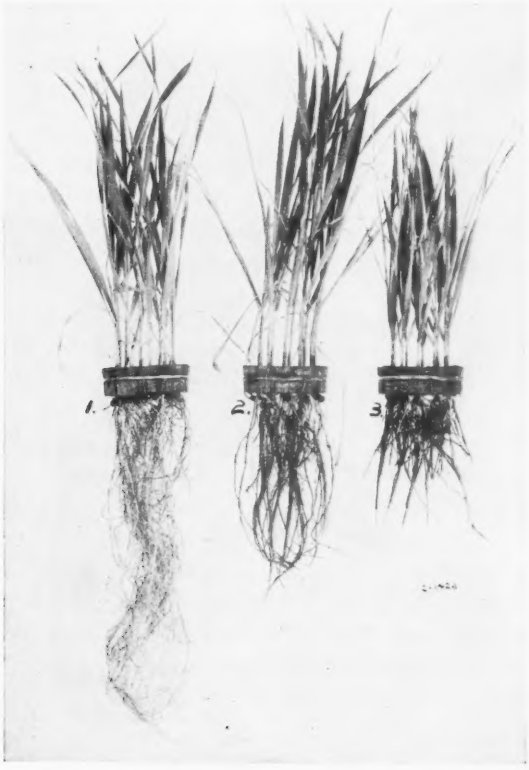

FIG. 12.—Wheat seedlings grown in water cultures: *1*, corn soil leachings; *2*, aerated kafir soil leachings; *3*, kafir soil leachings (no aeration).

The results of wheat grown in large containers of kafir and corn soil, as shown in table XI, present a marked varying in yield, even after calculating the probable error. Fig. 9 shows the extent of the variation in growth at an early stage of development. When

chopped up kafir roots were added to corn soil, the growth of wheat was depressed, compared with the growth in untreated corn soil, but the difference is not significant when the probable error is figured. These results are presented in table XII. In this experiment the kafir roots dried in the process of grinding them, which permitted the possibility of oxidation.

It has been pointed out from the work of PICKERING (12) that the toxic decomposition products of plants are quickly oxidized when exposed to the air. According to this idea, the results

FIG. 9.—Wheat growing in soil taken from surface seven inches of kafir plot (left), and from a corn plot (right).

obtained by the water culture methods of studying toxicity of soil solutions would not be reliable. On the other hand, this is the method used by SCHREINER and his co-workers (16), whereby it was found that the productive qualities of a soil were transmitted to its water solution. The arrangement of pot cultures with PICKERING trays, however, should demonstrate the comparative effects of one plant upon another. In table XIII, the results obtained with the employment of PICKERING trays are tabulated. There were five repetitions of each treatment. The corn and kafir

were grown in the trays placed in the jars filled with soil, and wheat was planted in the open holes. The wheat was grown to maturity and harvested. When the wheat grown in the case of solid bottom trays is compared, there is no difference in yield where corn or

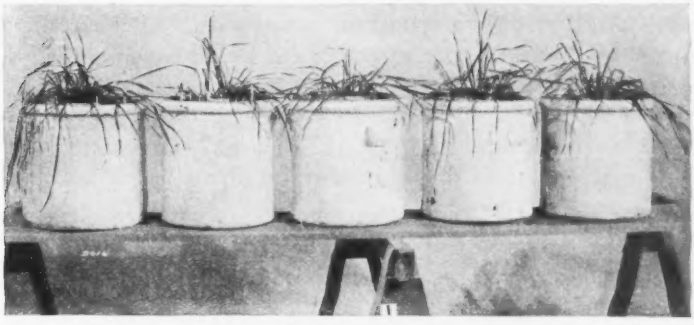

FIG. 10.—Pickering trays with perforated bottom; kafir produced in tray and followed by wheat in jar.

FIG. 11.—Pickering trays with perforated bottom; corn produced in tray and followed by wheat in jar.

kafir was the crop in the tray, but when a comparison is made of the perforated trays, kafir in the tray caused a depression of the wheat yield compared with corn in the tray. From this experiment it may be concluded that leachings from a soil producing kafir plants inhibits the growth of wheat as compared with the effect of corn plants.

In the summer of 1921, corn and kafir plants were grown in the trays placed in jars filled with soil, out in an open plant house. In the fall the plants were harvested, the jars were brought into the greenhouse, and wheat was planted in the open holes. The resulting growth during the early stages of development is shown in figs. 10 and 11. Here again kafir caused a depressing effect upon the yield of wheat. This effect was due to the leachings from the perforated trays.

Summary

1. Winter wheat grown after a kafir crop has yielded three bushels less per acre per year over a period of six years than wheat grown after corn. There is therefore an apparent depressing effect of kafir on the succeeding yield of wheat.

2. The kafir crop has produced 4000 pounds more of field cured dry matter than corn.

3. The kafir crop removes more fertilizing elements from the soil than corn, but this does not explain the effect of kafir in depressing the yield of wheat.

4. The growth of kafir does not affect the ability of the soil to liberate nitrogen.

5. The amount of moisture in the surface foot of soil is slightly lowered by the growth of kafir as compared with corn, which might influence the early growth of wheat in the field; but when this factor is under greenhouse control, the kafir soil does not produce as well as corn soil, showing that the depressing effect is not due to moisture differences.

6. Water culture studies conducted after the method of SCHREINER and SKINNER did not give evidence of any toxicity in kafir soil solutions, but because of the chance for oxidation of any possible toxic substances, these results cannot be taken as determining that there is not any toxicity in the soil as the result of the kafir crop. This seems the more certain in the light of the uncompleted experiments in progress, where leachings have been secured by an improved method, and in which wheat seedlings have been inhibited in root growth when placed in kafir soil leachings.

7. In pot cultures, where wheat received its moisture solely from water percolating through perforated trays in which kafir and corn

were grown, the growth of kafir plants inhibited the development of the wheat.

8. When kafir and corn were grown in these percolating trays, harvested, and then wheat planted where it would receive the drainage from the trays, the kafir soil trays depressed the growth of wheat. Consequently there are decomposition products from the crop residue which influence the growth of wheat.

Acknowledgments are accorded to Dr. WILLIAM CROCKER, Dr. CHARLES A. SHULL, University of Chicago, and to Dr. EDWIN C. MILLER, Kansas State Agricultural College, for their suggestions and counsel in the conduct of these investigations.

STATE AGRICULTURAL COLLEGE
MANHATTAN, KAN.

LITERATURE CITED

1. BENNETT, R. L., Soil improvement. Ark. Agric. Exp. Sta. Bull. 46:92–96. 1897.
2. BOLLEY, H. L., Flax wilt in North Dakota. N. D. Agric. Exp. Sta. Report 34–65. 1902.
3. ———, Causes of soil sickness in wheat lands. N. D. Agric. Exp. Sta. Bull. 107:96. 1913.
4. FLETCHER, F., Toxic excreta of plants. Jour. Agric. Sci. 4:245–247. 1912.
5. HALL, A. D., Theories of manure and fertilizer action. Science N.S. 28: 723, 617–628. *fig.* 1. 1908.
6. HARTWELL, B. L., and DAMON, S. C., The influence of crop plants on those which follow. R.I. Exp. Sta. Bull. 175. 1918.
7. HARTWELL, B. L., PEMBER, F. R., and MERKLE, G. E., The influence of crop plants on those which follow. R.I. Exp. Sta. Bull. 176. 1919.
8. HOLTER, G. L., and FIELDS, JOHN, Analyses of kafir corn fodder, whole plant and different parts. Same for corn fodder. Okla. Sta. Bull. 37. 17, 18. 1899.
9. JOHNSON, J., and HARTMAN, R. E., Influence of soil environment on the root rot of tobacco. Jour. Agric. Res. 17:41–86. 1919.
10. LIVINGSTON, B. E., BRITTON, J. C., and REID, F. R., Studies on the properties of an unproductive soil. U.S. Dept. Agric., Bur. Soils Bull. 28. 11–39. 1905.
11. PICKERING, S. U., and Duke of Bedford, Effect of one crop upon another. Jour. Agric. Sci. England 6:136–151. 1914.
12. PICKERING, S. U., The effect of grass on apple trees. Jour. Roy. Agric. Soc. England 64:365–376. London. 1903. Also 13th report of the Woburn Experimental Fruit Farm.

13. POTTER, R. S., and SNYDER, R. S., The determination of ammonia by the aeration method. Jour. Ind. and Chem. Eng. **7**:221. 1913.
14. RUSSELL, E. J., Soil protozoa and soil bacteria. Proc. Roy. Soc. (London) Ser. B. **89**:76–82. 1915.
15. RUSSELL, E. J., and PETHERBRIDGE, F. R., Jour. Agric. Sci. **5**:86–112. 1912.
16. SCHREINER, O., and SHOREY, E. C., The isolation of harmful organic substances from the soil. U.S. Dept. Agric., Bur. Soils Bull. 53. 53. 1909.
17. SCHREINER, O., and SKINNER, J. J., Some effects of harmful organic soil constituents. U.S. Dept. Agric., Bur. Soils Bull. 70. 98. 1910.
18. SCHREINER, O., and REED, H. S., BOT. GAZ. **45**:73–102. 1908.
19. SNYDER, W. P., and OSBORN, W. M., Rotation and tillage methods in western Nebraska. Neb. Sta. Bull. 155. 6–48. 1915.
20. TEN EYCK, A. M., and SHOESMITH, V. M., Indian corn. Kan. Exp. Sta. Report Bull. 147. 256–262. 1906–7.
21. VINALL, H. N., and GETTY, R. E., Growing and utilizing sorghums. Farmers' Bull. 1158. U.S. Dept. Agric. 1921.
22. WHITNEY, MILTON, and CAMERON, F. K., Investigations in soil fertility. U.S. Dept. Agric., Bur. Soils. Bull. 22. 1904.
23. WILLAMAN, J. J., WEST, R. M., SPRIESTERBACH, D. O., and HOLM, G. E., Notes on the composition of the sorghum plant. Jour. Agric. Res. **18**:1–31. 1919.

NOTES ON NEOTROPICAL ANT-PLANTS

II. TACHIGALIA PANICULATA AUBL.

I. W. BAILEY

(WITH PLATES I, II AND THREE FIGURES)

Introduction

In studying the myrmecophytes of the Kartabo region of British Guiana, Professor W. M. WHEELER and the writer became much interested in a plant with hollow, inflated petioles. Although we succeeded in tracing its development from germinating seeds to trees 20 m. high, we were unable to find flowering individuals. Thus we could not determine its systematic affinities until we returned to the United States and compared its vegetative characters with those of specimens in the Gray Herbarium of Harvard University. It proved to be a *Tachigalia*, but, owing to the fact that this leguminous genus is not well represented in American collections, we were obliged to send our specimens to Kew for comparison with the types of various Brazilian and Demararan species. Recently we received a letter from Colonel DAVID PRAIN, Director of the Kew Gardens, in which he states that our species is *T. paniculata* Aublet.

"Comparison has been made with AUBLET'S type of *T. paniculata* in the British Museum Herbarium, and there is no doubt that it is the same. The plant has been collected before in British Guiana by JENMAN, his specimen consisting of a large leaf only, which agrees with one of the samples communicated by you."

The plant

Tachigalia paniculata was described in 1775 by AUBLET, who derived the generic name from "tachigali," a word used by the Indians of the Guianas in referring to trees inhabited by stinging ants, "tachi," of the genus *Pseudomyrma*. A number of other myrmecophytic species of *Tachigalia* have since been described from various parts of neotropical America. Although these ant-plants are mentioned by SPRUCE (7) and other students of myrmeco-

phytism, they have never received the attention which they deserve, and little is known concerning the variability of their vegetative characters.

In view of the abundance of material of *T. paniculata* at our disposal, and its significance in taxonomic investigations, it is desirable to devote considerable attention to the description of plants of different ages and habitats. The species is fairly common

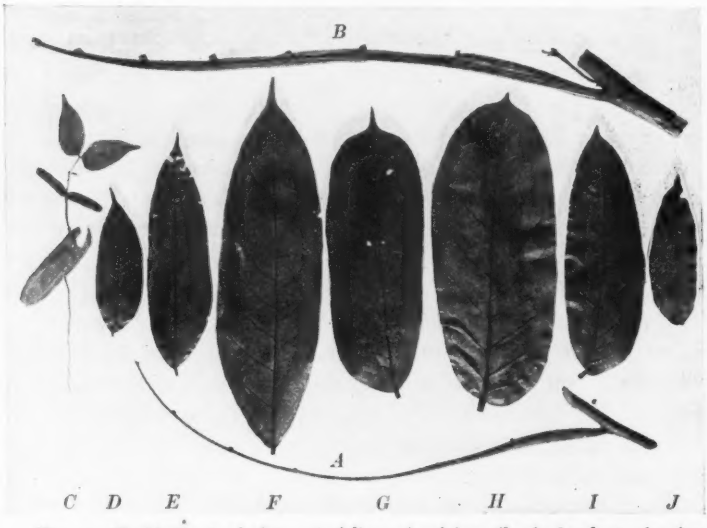

FIG. 1.—*Tachigalia paniculata: A*, foliar axis of juvenile shade plant, showing fusiform basal enlargement: *B*, foliar axis of vigorous sun plant showing fasciated character; *C*, indehiscent pod and seedling; *D*, leaflet from plant 4 dm. high; *E*, leaflet from shade plant 8 dm. high; *F*, leaflet from shade plant 3 m. high; *G*, leaflet from shade plant 6 m. high; *H*, leaflet from vigorous, rapidly growing sun plant; *I*, leaflet from slower growing sun plant; *J*, leaflet from base of floral axis of sun plant; $\times\frac{5}{16}$.

in the second growth of forests of the Kartabo region. The thin, indehiscent pods contain a single, flat, oblong seed which germinates readily in the leaf litter upon the forest floor (text fig. 1*C*). The primary leaf has two small, thin, glabrous, olive green, asymmetrical leaflets, and has two acicular, bifurcated stipules (text fig. 3*A*). As the seedlings develop, the leaves rapidly become larger, the leaflets increase in size and number (text fig. 1), and the stipules assume

a more complicated and foliaceous form (text fig. 3). The petioles and rhachises are slender, and resemble those of other young, woody Leguminosae. When the plants reach a height of 5–10 dm.,

FIG. 2.—*Tachigalia paniculata:* terminal shoot of vigorous, rapidly growing sun plant, showing fasciated appearance of cauline and foliar axes; $\times \frac{5}{32}$.

and the leaves a length of approximately 25 cm., the petiole and the basal portion of the rhachis tend to become inflated, forming a hollow, fusiform hypertrophy 5–8 cm. long (text fig. 1*A*). The

flat, upper surface of this spindle-shaped swelling is finely canaliculate, and the convex under surface is somewhat hexangular (figs. 1, 2).

At this stage of their development the plants take on a characteristic form and structure, which they tend to retain so long as they continue to grow in the feebly illuminated interior of the jungle. The long, slender, nearly terete, unbranched stems bear a few relatively large, widely separated, alternate leaves which have 12–16 thin, glabrous, dark olive green, nearly horizontal leaflets (text fig. 1*E–G*). The leaves increase in size until they attain a length of 60–80 cm. As the rhachis elongates and thickens, it tends to become somewhat triangular and slightly alate, and to develop a longer internal cavity. The basal inflation tapers gradually toward the apex of the rhachis, and is less conspicuous, therefore, than it is in seedling leaves where the transition is more abrupt.

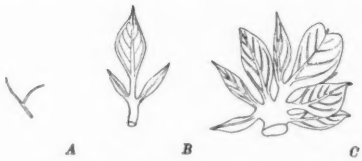

FIG. 3.—*Tachigalia paniculata* (stipules): *A*, from primary leaf of seedling; *B*, from juvenile shade plant; *C*, from vigorous sun plant; natural size.

When the saplings become exposed to direct sunlight, they begin a period of active growth and differentiation. The axillary buds form branches, and the stems and leaves undergo a striking metamorphosis. In fact, the vegetative characters of dominant trees are so unlike those of young suppressed individuals that it is difficult to realize that the two types of plants actually belong to a single species. The young shoots and leaves of typical adult sun plants are yellowish or reddish brown, whereas those of juvenile shade plants are dark olive green. The rhachises of the leaves of the former plants are short, stout, hollow, distinctly triangular, and deeply corrugated (text fig. 1*B*, figs. 6, 8, 9). The leaflets (text fig. 2) are thick, coriaceous, closely crowded, pendant, and conspicuously embossed-reticulate on the upper surface when dry, and the large foliaceous stipules are palmately multifid (text fig. 3*C*). There are 6–8 rufous, tomentose buds, arranged in a linear series, in the axil of each leaf. Furthermore, the large, asym-

metrical, deeply corrugated terminal shoots have a distinctly fasciated appearance, particularly during the earlier stages of their development (fig. 7).

The leaves and leaflets of the sun plants vary considerably in size and shape, as do those of the young shade plants (text fig. 1). The latter increase in size as a plant matures, whereas the former tend to do the reverse. The leaflets of seedlings, young saplings, and old, slow growing trees are lanceolate-acuminate, but the large leaflets of vigorous transitional types of plants frequently are oblong-mucronate and are obtuse at the base. It should be noted in this connection, however, that all of the leaflets, regardless of fluctuations in size, shape, color, and texture, tend to be more or less asymmetrical.

It is evident, accordingly, that in dealing with tropical plants, whose vegetative characters are extremely variable, the investigator should collect a series of typical specimens, particularly when the flowers are not available. Thus, the identity of the juvenile Tachigalias, in which we were specially interested, could not be determined without the traditional types of foliage. This is due, of course, to the fact that descriptions of species are based upon more or less fragmentary type specimens, and usually afford no clue to the range of variability of the various diagnostic characters. As in the case of the Cecropias, discussed in the first paper of this series,[1] the mature vegetative shoots and leaves of *T. paniculata* frequently are too large for herbarium sheets of standard dimensions, and the average botanist instinctively tends to collect the small foliar organs at the base of the floral axis.

Origin, structure, and significance of the foliar hypertrophies

SPRUCE[2] and BECCARI (3) believed that the fistulose stems and petioles, bullate leaves, hollow thorns, and other structural "peculiarities" of myrmecophytes are produced by the stimulating activities of ants. They were forced to admit, however, that such traumatisms ultimately may become inherited. BELT (4) and other

[1] BOT. GAZ. 74:369–391. 1922.

[2] SPRUCE read his paper before the Linnean Society in 1869, but it was not published until 1908.

advocates of the theory of myrmecophily argued that the structures are adaptations for attracting a defending army of ants, and have been acquired presumably through the action of natural selection. More recently, CHODAT and CARISSO (6) have formulated "une nouvelle théorie de la myrmécophile," in which they assert that the hypertrophied organs of ant-plants are in reality galls which become inhabited by ants after they are abandoned by gall forming insects.

Although ants may produce slight alterations in the hollow or inflated axes of myrmecophytes, and gall insects may oviposit in them, there is no reliable evidence to indicate that the structural "peculiarities" of ant-plants actually are initiated by these insects. In *T. paniculata*, as in the numerous Ethiopian ant-plants investigated by BEQUAERT (5) and the writer (2), the myrmecodomatia are not inhabited by gall insects during the earlier stages of their development, and are not colonized by ants until they have nearly attained their maximum size. Furthermore, the localized, fusiform hypertrophies of juvenile Tachigalias must be correlated with the hollow petioles and rhachises of adult sun plants. Although the former have a slight superficial resemblance to certain types of galls, the latter obviously are normal structures. As the leaf elongates and increases in weight during the ontogeny of the plant, the enlargement of the foliar axis begins at its base and gradually extends toward its apex.

That the fistulose stems of *Cecropia*, *Triplaris*, and of other ant-plants are not adaptations for housing ants, and may be explained on the mechanical principle of construction as being the method of producing the greatest resistance to bending with the least expenditure of material, was admitted by SCHIMPER, the ablest advocate of myrmecophily. The triangular or carinate foliar axis of *Tachigalia*, as of other plants which are not inhabited by ants, is a simple modification of the ordinary cylindrical type. It is an extremely rigid structure, and one which provides favorable attachments for the two rows of relatively heavy leaflets. The internal cavity, in all probability, is merely a concomitant of the excessive enlargement of the pith, for we have found that unusually large and vigorous cauline axes (fig. 7) tend to become fistulose.

In addition, it is to be emphasized that plants with compound leaves and relatively large leaf traces frequently have angled or corrugated stems, petioles, and rhachises. In *T. paniculata*, as in many other Leguminosae, the leaf traces (three to each leaf) are set off some distance below the node, and produce prominent ridges upon the surface of immature stems (figs. 3, 7). This is due to the fact that the foliar strands, which are the first formed portions of the fibrovascular cylinder, are larger than the adjacent bundles of the stele. As the three leaf traces pass into the petiole they divide, anastomose, and form a foliar siphonostele which is not unlike that of the cauline axis. It is composed of strands of different sizes which are separated by gaps, that is, primary medullary rays of varying widths (fig. 8). The fasciated appearance of the immature foliar axes of vigorous sun plants is due to the fact that there are as many ridges and grooves on their surfaces as there are bundles and gaps in their steles. With the initiation of a cambium, which tends to bridge the intervals between the separate bundles, many of these irregularities may ultimately be smoothed out. In the inflated basal portion of the foliar axis, certain of the wider gaps in the fibrovascular cylinder are not closed by the formation of secondary wood, but tend to remain permanently parenchymatous. There are four of these wide primary medullary rays in the petioles of juvenile plants (fig. 1), and several of them in those of sun plants (figs. 6, 8, 9). They tend to increase in depth (radial) as the walls of the petioles become thicker, and their cells are filled with an amorphous, amber colored substance, as are those of the peripheral layer of parenchyma which jackets the medullary cavity (fig. 5).

Activities of ants and other insects

Insects are so abundant in the moister regions of the tropics, that there is a very keen competition for nesting places. Any easily accessible cavity of suitable dimensions which provides a safe refuge is quickly discovered and utilized. It is not surprising, therefore, to find that fierce conflicts are waged for possession of the hollow foliar axes of *Tachigalia*. WHEELER (8) and the writer found more than thirty different species of insects in the inflated petioles of

juvenile plants. Seven of these are definitely attached to *Tachigalia* as their host plants, and may be designated as "obligatory," whereas most of the others are "facultative," that is, inquilines which take possession of old domatia that have been abandoned by previous tenants. The former include *Pseudomyrma damnosa* Wheeler, *P. maligna* Wheeler and its two varieties *cholerica* Wheeler and *crucians* Wheeler, *Azteca foveiceps* Wheeler, and two extraordinary colonial beetles, to which SCHWARZ and BARBER have assigned the names *Coccidotrophus socialis* and *Eunausibius wheeleri*. The latter are confined to seedlings and young saplings, since the older plants are dominated by one of the five obligatory ants.

The beetles enter their domatium through an oval opening cut in one of the thinner sides of the fusiform hypertrophy. The dried remains of the succulent inner portion of the pith are trimmed away, and are packed into the ends of the spindle-shaped cavity. The beetles then excavate and feed upon the peripheral layer of amber colored medullary tissue, and the similarly colored parenchyma which fills the wide gaps in the fibrovascular cylinder (figs. 1, 2, 4). As this tissue is consumed, the resulting frass is piled in parallel ridges, and the larvae and imagines move backward and forward in the intervening grooves. Most petioles which contain flourishing colonies of beetles also contain numerous coccids, *Pseudococcus bromeliae* Bouché, which feed upon the amber colored substance in the parenchyma of the primary medullary rays. Both the larval and the imaginal beetles solicit and obtain sugary exudations from the coccids by stroking them with their antennae. Thus the beetles are able to feed both directly and vicariously upon the softer tissues of the plant. In the course of time, the wide rays are cut back nearly to the level of the cortex (fig. 2). At pupation the larvae construct their puparia from freshly excavated bits of this nutritive parenchyma (fig. 2).

The inflated petioles of juvenile Tachigalias frequently are entered by young *Azteca* queens, but they usually are killed by beetles before they can initiate a colony. During the subsequent stages in the development of the plants, and as the leaves increase in number, a queen ant sooner or later succeeds in raising a brood in a petiole

which is not inhabited or visited by beetles. As soon as this happens, the numerous worker ants swarm over the plant, kill or drive out the beetles, and take possession of all of the domatia. If the latter contain coccids, they become the spoil of the victors. As the ants increase in number, portions of the colony migrate to the hollow foliar axes of the successively formed leaves, which are used as additional dwellings or brood chambers. The workers cut one or more circular entrances in the thinner sides of the petioles, and remove the desiccated portions of the pith. They then cut back the peripheral layer of medullary tissue and transport part of the brood to the new domatium.

In excavating the tissue which jackets the petiolar cavity, the Pseudomyrmas cut into the inner surface of the primary rays, thus producing a series of long, narrow, shallow grooves (fig. 6). The stimulus produced by the gnawing of the ants accelerates the formation of amber colored parenchyma, leads to the widening of the primary rays, and may even inhibit the formation of thick-walled elements in portions of the foliar axis which otherwise would become woody. The Aztecas, in addition, excavate pits in the walls of the petiole and rhachis. Many of these excavations are so deep that they perforate the epidermis, and later have to be blocked up with compact masses of triturated parenchyma (fig. 9). In fact, it is possible to determine whether a given tree is inhabited by Aztecas or by Pseudomyrmas by the presence or absence of these numerous occluded perforations. The softer tissues on the margins of the wounds proliferate and form callus. The large, thin-walled cells of this traumatic parenchyma are filled with amorphous, amber colored contents, therefore resembling those of the primary rays.

As soon as the foliar axes become inhabited by ants, they are found to contain coccids, *Pseudococcus bromeliae* Bouché or occasionally *Ripersia petiolicola* Morrison, which feed upon the amber colored parenchyma of the wide primary rays, and, in the case of plants inhabited by Aztecas, also upon the amber colored traumatic tissue in the pitlike excavations. The coccids are more numerous in domatia inhabited by Aztecas than they are in those which are colonized by Pseudomyrmas.

Discussion

That the beetles feed directly upon the amber colored parenchyma of the primary medullary rays is shown by analyses of their faeces or of the contents of their alimentary tracts. Unfortunately, it is not possible to secure conclusive evidence in regard to the feeding habits of the Aztecas and Pseudomyrmas by similar methods. This is due to the fact that the crops and stomachs of most ants, and of all imaginal forms, contain only a structureless mush of liquid or semiliquid substances. Fragments of plant or of animal tissues, removed by means of the mandibles, are not swallowed *in toto*, but their more fluid or easily soluble portions are removed by the roughened surfaces of the mouth parts. I have shown in a previous paper (1) that the so-called infrabuccal pouch merely serves as a repository for detritus, and therefore is not significant in this connection. Only in the case of the Pseudomyrminae, in which the larvae are provided with a curious pouch or trophothylax which is periodically filled with food by the workers, is it possible to secure evidence in regard to the food by microscopic analyses. Furthermore, to argue, as many investigators do, that tissues which are excavated by the ants actually are eaten by these insects is extremely misleading, since such tissues may be removed for various reasons, for example, in cleaning and smoothing the walls of the domatia, for the construction of carton, to prevent or hold in check the growth of obnoxious fungi, for facilitating the feeding of coccids, etc.

That the ants do not feed extensively upon the amber colored parenchyma is indicated, however, by the fact that in domatia which are crowded with larvae and imagines (figs. 6, 9), the strands of this tissue are not gnawed back as they are in petioles inhabited by relatively small colonies of beetles (fig. 2). Indeed, it seems probable that the ants do not feed directly upon the ray tissue, but only vicariously through the intervention of coccids. Such an assumption is supported by the activities of ants in other myrmecophytes. In the first paper of this series, I showed that in *Cecropia* the Aztecas excavate small pits in the horny layer which jackets the internodal chambers, and thus enable the coccids to insert their setae into the softer underlying tissue. Attention has also

been called to the fact that in Ethiopian myrmecophytic species of *Cuviera*, *Plectronia*, *Barteria*, and *Sarcocephalus*, the ants cut into the thinner walls of their domatia, as far as the cambium or cortex, and induce the formation of a "nutritive" callus which is fed upon by coccids. Only in *Vitex Staudtii* Guerke, in which the highly specialized *Viticicola tessmanni* Wheeler feeds upon the callus, is this layer cut back to a depth corresponding to that in beetle-inhabited Tachigalias; and the Viticicolas do not tend coccids.

The beetles and Aztecas solicit and obtain liquid carbohydrates from the coccids by stroking them with their antennae, but the Pseudomyrmas belong to a group of ants which in general do not devote much attention to coccids, aphids, membracids, and other sugar excreting insects. During our limited residence at Kartabo, we did not succeed in perfecting methods for studying the feeding habits of these fiercely stinging and extremely active ants in artificial nests. In the case of the nearly related African Pachysimas, the writer succeeded in demonstrating, by analyses of the pellets which are fed to the larvae, that the ants carve up and eat the coccids. In other words, they use them for beef rather than solely as miniature milch cows. Analyses of the larval pellets of the *Tachigalia*-inhabiting Pseudomyrmas reveals the fact that these ants also eat the coccids, or rather feed them to their larvae. It is significant in this connection that, although the coccids are thereby considerably reduced in number, they never, so far as we have been able to determine, are entirely exterminated. A sufficient number of adults survives for replenishing the herd.

It is evident, accordingly, that the obligatory guests of *Tachigalia paniculata*, both the ants and the beetles, are dependent either directly or indirectly upon the plant for a considerable portion of their food. Their relations to their hosts, therefore, are entirely different from those of the numerous facultative inquilines which utilize the hollow foliar axes merely as convenient nesting chambers. What then is their significance in a discussion of the theory of myrmecophily? According to this hypothesis, the structural peculiarities of *Tachigalia* should be interpreted as adaptations for attracting an army of ants which protect the plant against the

attacks of phytophagous animals, particularly the leaf cutting, Attine ants. Although *T. paniculata* is not provided with numerous extra-floral nectaries, food bodies, or preformed entrances (prostomata), the inflated petioles and the strands of "nutritive" parenchyma might be cited as specializations acquired through the action of natural selection. It should be noted, however, that during the earlier stages of their development, the plants are not dominated by the Aztecas or by the Pseudomyrmas. The petioles are inhabited by beetles which remain within their domatia and obviously do not protect their hosts. Of course it may be argued that juvenile plants do not need the protection of the ants, but WHEELER found small saplings which had been defoliated by Attas. Furthermore, the leaf cutting ants show no particular preference for the foliage of adult Tachigalias, as the writer determined by placing fresh leaves in close proximity to a large nest of *Atta cephalotes* L. The ants did not molest the tough, leathery leaves, although they did cut into the foliage of other plants which were used as controls. We could find no evidence to indicate that *Tachigalia* needs the protection of the Aztecas or Pseudomyrmas, or that such protection would be afforded if actually needed, nor are there logical arguments for assuming that the hollow foliar axes and the strands of amber colored parenchyma are allurements for attracting ants. Fistulose axes and wide primary medullary rays occur normally in many plants which are not visited by ants, and their presence may satisfactorily be accounted for as due to necessary anatomical, physiological, and mechanical adjustments.

The relations between the Tachigalias and their various guests do not appear to be those of a mutually beneficial symbiosis, but purely one of parasitism, in which the insects avail themselves of the structural peculiarities of the plants in a singularly efficient manner. That representatives of two such widely separated groups of insects as the Hymenoptera and the Coleoptera should discover and utilize the hollow foliar axes of *Tachigalia* is not surprising, in any region where the competition for suitable nesting places is as keen as it is in the tropics, but that they should develop such complex and delicately adjusted relations with coccids is indeed significant from the point of view of insect behavior.

In conclusion, it is to be emphasized that in studying the habits of phytophagous and of plant inhabiting insects, the anatomy of the host plants deserves more careful attention than it has received heretofore. In the case of myrmecophytic plants, the nesting and feeding habits of the ants, and their relations to coccids and fungi, are largely determined by the structure and arrangement of the various vegetative tissues during different stages in the development of the stem or of its various appendages.

Summary and conclusions

The hollow foliar axes of the ant-plant *Tachigalia paniculata* are colonized by at least seven "obligatory" guest insects. The domatia of juvenile plants are taken possession of by two extraordinary social beetles, but during the subsequent development of the plants these insects are dispossessed by ants. Both the beetles and the ants avail themselves of the structural peculiarities of their host plant in a singularly efficient manner. Not only do they make use of the hollow foliar axes as convenient nesting chambers, but they feed, either directly or indirectly, upon the softer tissues in the interior of the domatia. The beetles eat the parenchyma of the wide, primary medullary rays, utilize it in the construction of their puparia, and solicit and obtain liquid carbohydrates from herds of coccids which graze upon it. The ants also feed upon this tissue vicariously through the intervention of coccids, and utilize it in the construction of carton partitions.

There is no evidence to indicate that the structural peculiarities of *T. paniculata* are initiated by ants or by gall forming insects, or that they originated as adaptations for attracting a defending army of ants. The relations between the host plant and the beetles and the ants are not those of a mutually beneficial symbiosis (BELT, SCHIMPER), but an interesting type of parasitism, in which there is a remarkable parallelism in the behavior of representatives of such widely separated groups of insects as the Hymenoptera and the Coleoptera. The nesting and the feeding habits of the insects, and their relations to the coccids, are largely determined by the structure and arrangement of the various vegetative tissues during different stages in the development of the petiole and rhachis.

In studying the habits of phytophagous and plant inhabiting insects, it is evident that the anatomy of the host plants deserves more careful consideration than it has received heretofore.

This investigation was conducted under a grant from the American Association for the Advancement of Science. I wish to thank WILLIAM BEEBE for his kindness in facilitating my investigation during my visit to the Tropical Station of which he is in charge. To Professor B. L. ROBINSON of the Gray Herbarium of Harvard University, and to the Director of the Kew Gardens, I am indebted for notes and comments upon various species of *Tachigalia*.

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LITERATURE CITED

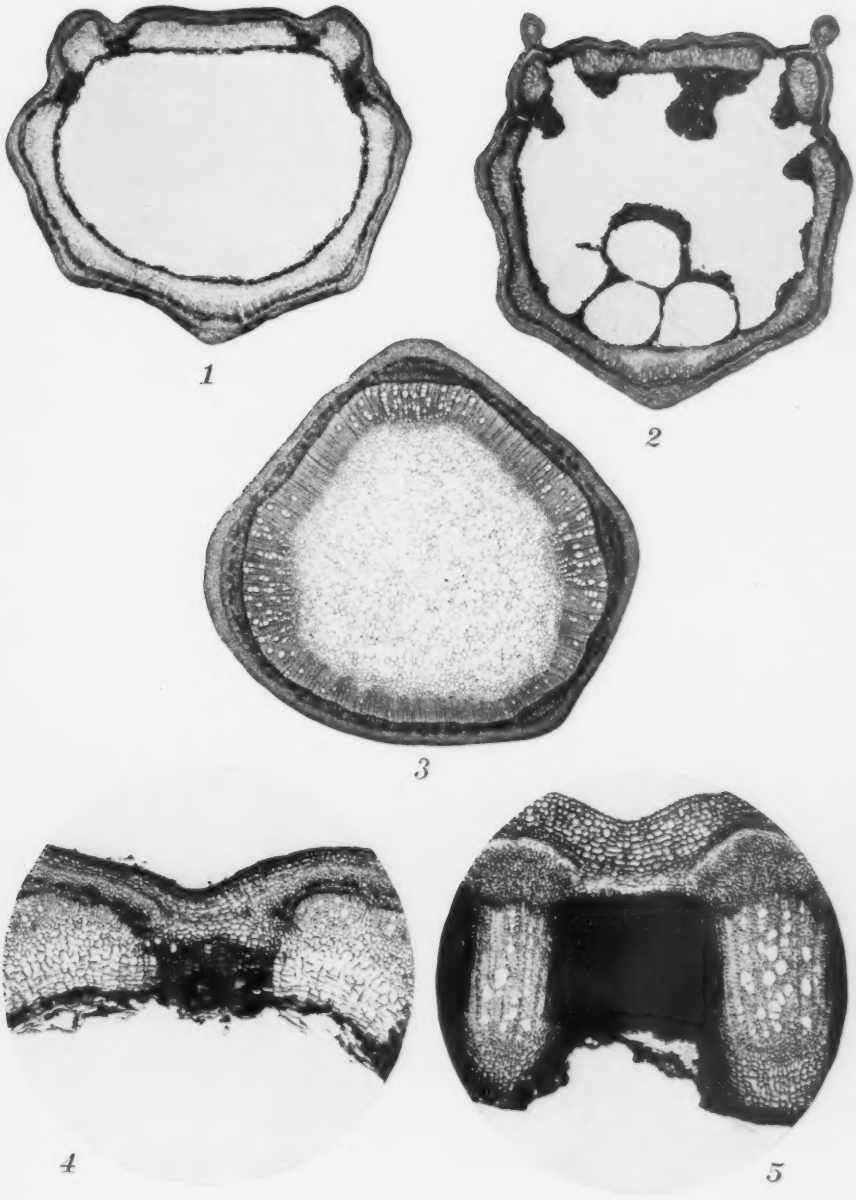
1. BAILEY, I. W., Some relations between ants and fungi. Ecology **1**:174–189. 1920.
2. ———, The anatomy of certain plants from the Belgian Congo, with special reference to myrmecophytism. Bull. Amer. Mus. Nat. Hist. **45**:585–621. 1922.
3. BECCARI, O., Piante ospitatrici ossia piante formicarie della Malesia e della Papuasia. Malesia **2**:1–340. 1884–86.
4. BELT, T., The naturalist in Nicaragua. London. xvi+403. 1874.
5. BEQUAERT, J., Bull. Amer. Mus. Nat. Hist. **45**:333–583. 1922.
6. CHODAT, R., and CARISSO, L., Une nouvelle théorie de la myrmécophilie. Compt. Rend. Soc. Phys. Hist. Nat. Genève **37**:9–12. 1920.
7. SPRUCE, R., Notes of a botanist on the Amazon and Andes. **2**:xii+542. 1908. Edited by A. R. WALLACE.
8. WHEELER, W. M., A study of some social beetles in British Guiana and of their relations to the ant-plant *Tachigalia*. Zoologica **3**:35–126; The *Tachigalia* ants. *Ibid.* 137–168. 1921.

EXPLANATION OF PLATES I, II

FIG. 1.—*T. paniculata:* transverse section of petiole of juvenile plant, showing medullary cavity and four strands of amber colored parenchyma (black); from foliar axis which had not been colonized by ants or by beetles; ×12.

FIG 2.—Transverse section of petiole of juvenile plant, colonized by beetles, showing puparia and frass ridges; primary rays cut back nearly to level of cortex; ×8.

FIG. 3.—Transverse section of stem of juvenile shade plant; ×10.



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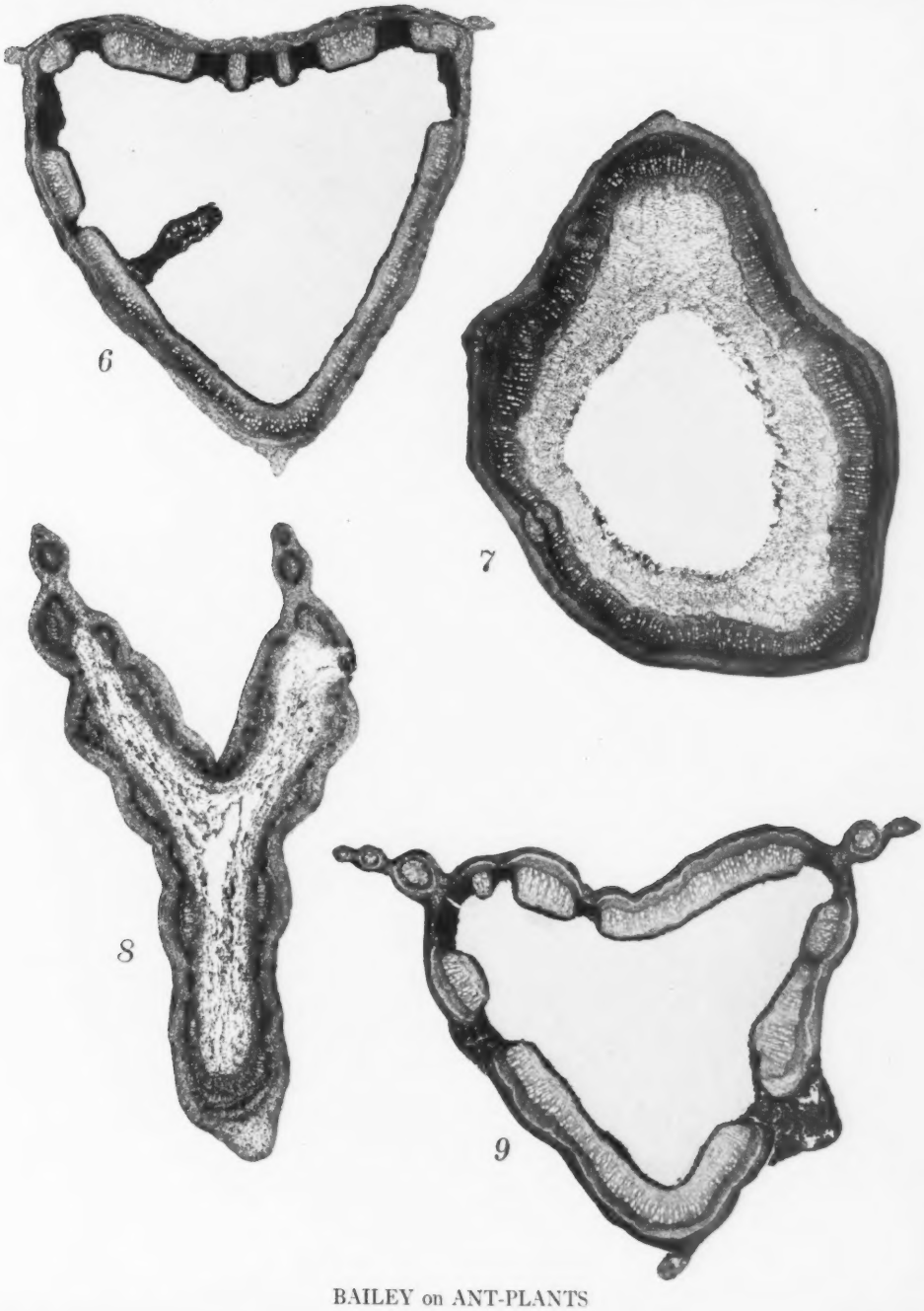
BAILEY on ANT-PLANTS



BAILEY on ANT-PLANTS

FIG. 4.—Portion of fig. 1, more highly magnified, showing strand of amber colored parenchyma; ×35.

FIG. 5.—Portion of fig. 6, more highly magnified, showing wide, primary medullary ray; ×44.

FIG. 6.—Transverse section of petiole of vigorous sun plant, showing medullary cavity, primary rays (black), and portion of carton partition; from plant colonized by *Pseudomyrma maligna;* ×6.

FIG. 7.—Transverse section of stem of vigorous sun plant, showing fasciated character and medullary cavity; ×6.

FIG. 8.—Transverse section of immature foliar axis of vigorous sun plant, showing formation of medullary cavity and many bundled stele; ×10.

FIG. 9.—Transverse section of petiole of vigorous sun plant, showing primary rays and perforations blocked with triturated parenchyma; from tree colonized by *Azteca foveiceps;* ×7.

DIFFERENTIATION AMONG CHROMOSOMES IN ELODEA

CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY 302

JOSE K. SANTOS

(WITH PLATE III)

A problem as important as the determination of sex always suggests further investigation, and since *Elodea* is rather favorable for a study of the chromosomes, on account of its structure and behavior, an investigation was undertaken at the suggestion of Professor CHARLES J. CHAMBERLAIN, to whom the writer is indebted for criticism, advice, and for some of the material.

STRASBURGER (31), in reviewing the theory that sex is determined by surrounding conditions, such as food, heat, light, or other external factors, concluded that the evidence was not sufficient. His own experiments with dioecious forms indicated that sex is not determined by environmental factors, but is inherent in the germ cells.

Cytological work on the determination of sex in plants is just beginning, the principal papers being those of DARLING (6) on *Acer Negundo*, in which he reviewed the literature up to 1909, and the critical work of ALLEN (1) on *Sphaerocarpus Donnellii*. Work on animals is much more extensive, and some very decisive results have been obtained.

DARLING concluded that the meiotic mitoses in *Acer Negundo* are somewhat analogous to those in some insects. The fact, however, that the division of the chromosomes at the second mitosis could not be determined with certainty would indicate that the technique was hardly sufficient to establish the claim that the eight chromosomes are formed by the segmentation of the spireme thread, and that the five bivalent chromosomes come out bodily from the nucleolus.

MOTTIER (23), working on *Acer Negundo* pollen mother cells, got results differing from those of DARLING, for he found that in

the resting stage of pollen mother cells the chromatin is distributed upon a linin network, and not contained in the nucleolus. Before synapsis there is no chromatin spireme, the spireme being formed after synapsis by a serial arrangement of chromatin granules. All the chromosomes are formed by segmentation, and not by budding off bodily from the nucleolus. The chromatin is equally distributed to the four microspores, so that their nuclei are all alike.

No other work up to the present has done more to throw light upon the sex question in plants than ALLEN's (**2**) carefully conducted experimental and cytological investigations on dioecious Bryophytes. The results obtained in his experiments with *Sphaerocarpus Donnellii* are practically in accord with those obtained by C. and R. DOUIN (**10**), who worked with the same genus. There are two kinds of spores, half of which have a tendency to become female and half male. These results are also in accord with those of the MARCHALS (**16, 17**) on mosses, CORRENS (**5**) on *Bryonia dioica* and *B. alba*, NOLL (**25**) on *Cannabis*, STRASBURGER on *Mercurialis annua* and *Melandrium album*, and others. In the cytological study, ALLEN found that the chromosome group in the cells of the female gametophyte of *Sphaerocarpus Donnellii* contains one large element which considerably exceeds both in length and in thickness any of the other chromosomes. The chromosome group of the male gametophyte contains no element similarly distinguished by its size; but it has a very small chromosome which does not seem to correspond in size to any element in the female. These results show that, in consequence of the chromosome distribution in the reduction divisions, two of the four spores derived from a single spore mother cell receive each a large chromosome and seven smaller ones, and that these spores develop into female plants; while each of the other two spores receives a small chromosome instead of the large one, and, on germination, gives rise to a male plant. On account of the resemblance between this behavior and that of the chromosomes of certain insects, such as *Lygaeus* and *Euschistus*, which possess a large *X* and *Y* chromosome, ALLEN came to the conclusion that these particular chromosomes of *Sphaerocarpus* are the bearers of definite sex determining factors.

Material and methods

Among the Angiosperms the genus *Elodea* is exceptionally favorable for the study of sex in plants, since it is strictly dioecious, and in *E. canadensis* the four spores of the tetrads cling together even in the shedding condition. This induced STRASBURGER (**32**) to begin a study, which unfortunately was only in its initial stages when he died. He had pollinated hundreds of stigmas, each with a single tetrad, and had anticipated that four eggs would be fertilized, and that two of the resulting seeds would produce male plants and two female. This laboriously prepared material was not investigated. The writer, at the suggestion of Professor CHAMBERLAIN, undertook a cytological study of *Elodea*, and in August 1921 collected material from Wolf Lake, about fourteen miles from Chicago, where STRASBURGER'S material had been secured. As the flowering season was over, a quantity of the plant was placed in the greenhouse of the University of Chicago. Near the end of September male flowers appeared, but no female. Material was fixed in chromacetic acid, with about ten drops of osmic acid to 50 cc. of the solution. When the flowers were examined, they were a little too old, only two or three showing the desired stages. *Elodea gigantea* was available, having been collected a couple of years earlier, and this material was kindly furnished, ready for imbedding.

The principal difference between the two species is that *E. gigantea* is more vigorous, with broader and longer leaves, and with tetrads which do not cling together like those of *E. canadensis*. As the purpose of the present investigation was to study the chromosome differences in relation to sex determination, it was immaterial whether the tetrads cling together or not. The material used was cut 3–5 μ thick, with some 10–15 μ, and Haidenhain's iron-alum haematoxylin was used for staining.

Resting stage to synizesis

The pollen mother cells, just after they are separated from the tapetal cells, have a polygonal outline in both transverse and longitudinal section of the anther. They are easily distinguished from the cells from which they were formed, by being filled with a denser, finely granular cytoplasm, and by the fact that their nucleus soon

becomes very large. The nuclei remain in resting condition for a short period only. They contain generally a single big and vacuolated nucleolus, in a more or less eccentric position in the delicate and scanty chromatin network. This threadwork contains nodelike structures or enlargements like beads in the intersections of the threads. There is no trace of prochromosomes in the resting stage, but there is evidence that the spaces in the network are due to the pulling apart of the sides of the chromosomes where fission has taken place.

The first evidence that synizesis is about to take place is the appearance of a clear space at one side of the nucleus, the thickening of the chromatin threads or meshes, and the enlargement of the nodelike beads of chromatin. Perhaps a partial precipitation of the nuclear sap, due to the fixing of the material, has some relation to this enlargement of the chromatin nodes. If the chromatin threads are examined carefully at this stage, they appear doubled, and the chromatin nodes seem to be in pairs. This double nature suggests that the apparent thickening of the chromatin threads is partly due to the individual threads, which are brought close together by contraction. The same thing is observed with the nodes. During this period of development of the nucleus there are two distinct and interesting phenomena, the apparent contraction of the reticulum and the enlargement of the nuclear cavity. This contraction of the nuclear contents, so long described by cytologists, is denied by LAWSON (**15**), who believes that the stage, commonly known as synapsis, is simply due to a sudden growth of the nucleus, and not to the contraction of the reticulum. On the other hand, DAVIS (**7**) suggested that contraction is due to shortening and thickening of the threads of the network previous to their transformation into spireme. In view of these statements, the writer made a series of measurements, starting with the resting stage and ending with synizesis. These measurements show conclusively that in *Elodea* there is a contraction of the reticulum as well as a growth of the nucleus. GATES and REES (**13**), from a series of measurements in the growth period of *Lactuca* pollen mother cells, concluded that in synizesis the reticulum undergoes contraction to approximately half the diameter of the resting nucleus, and that there is

steady growth of the nucleus rather than the sudden growth described by LAWSON (15).

If the cause of this apparent contraction is imperfect fixation of the material, as claimed by LAWSON, perhaps the only possible way to clear up the matter is to study and measure the different stages of the growth period of the nucleus in the living condition. As far as the fixed material is concerned, however, there is conclusive evidence that a contraction of the reticulum and a growth of the nucleus occur simultaneously. Measurements of a great many nuclei of different stages, and cut in a median plane through the chromatin mass, are given in table I.

TABLE I

DIMENSIONS OF NUCLEI IN *Elodea gigantea*

RESTING STAGE	
Largest	12.2 × 9.3 μ
Smallest	9.6 × 7.6 μ
Average	11.7 × 8.9 μ
EARLY SYNIZESIS	
Average	17.6 × 12.7 μ
SYNIZESIS	
Nucleus	
Largest	23.94 × 18.62 μ
Smallest	14.63 × 13.3 μ
Average	18.75 × 16.5 μ
Reticulum	
Largest	9.31 × 7.9 μ
Smallest	6.65 × 5.32 μ
Average	8.1 × 6.6 μ

Most of the averages are calculated from forty measurements, but some from only twenty-five measurements. By comparing these figures it is evident that in synizesis contraction proceeds to approximately one-third the diameter of the resting nucleus. While the contraction is in progress the chromatin threads become increasingly distinct, and the nodes become closer and more definitely arranged. Sometimes fine threads in bundles are observed bridging the space between the nuclear membrane and the reticulum. This contraction proceeds until at last a compact ball of chromatin threads at one side of the nucleus is gradually formed. Generally, but not

always, the nucleolus is inclosed and squeezed by the tight knot of threads. This synizesic ball, formerly called synaptic ball, remains for a comparatively long period, but the exact duration of it or the condition regulating it was not determined. In this stage the chromatin bodies become almost indistinguishable from the reticulum, and the chromatin threads of the latter become gradually rearranged from an anastomosing reticulum to a long continuous double or paired thread. The exact nature of this rearrangement has not yet been discovered.

Soon after the synizesic contraction the knot loosens its structure, and the closely doubled threads begin to extend gradually throughout the nuclear cavity until they are uniformly distributed in it. In the meanwhile the spireme becomes thicker, and appears as a homogeneous, smooth, single, and continuous filament, except that in some places free ends may be observed. Immediately after the distribution of the spireme in the nuclear cavity beautiful loops are formed, which are especially characteristic before the second contraction takes place.

Shortly after the numerous loops are formed and spread through the nuclear vacuole, the spireme passes gradually into the condition known as the second contraction, characterized by the thickening and shortening of the chromatin filament. The loops become rearranged, their sides begin to approach each other, and they gradually become detached. This is the most critical period of the nuclear development. The segments of the spireme become entangled, and it is almost impossible to determine the nature and number of the individual pieces. After a careful study of various nuclei in this stage, however, the writer believes that the single segments unite end to end, and occasionally some of them may be found twisted about each other.

Diakinesis

The most striking feature of this stage is the partial condensation of the young heterotypic chromosomes. These in pairs become individualized and acquire a more or less definite form. They appear rod-shaped, of various lengths, lying side by side, and in some cases slightly twisted about each other. Fig. 1 represents a typical view of the chromosome at diakinesis stage, in which one of

the pairs is conspicuously wider and longer, while another pair is conspicuous on account of the unequal length of its two members. The large chromosome, which the writer might call the giant chromosome, is distinctly double. This is noticed even when the maximum condensation has been attained by the chromosome.

In the late diakinesis stage, the nuclear membrane is extremely thin and the nucleolus gradually decreases in size, and in some instances looks as if it were being reduced into fragments. At the time the spindle fibers appear, the nuclear membrane and the nucleolus disappear simultaneously. This has led some to suggest that both the membrane and the nucleolus have been used in the formation of this spireme, or that in some way or other they contribute to the formation of spindle fibers. This was especially emphasized by HARPER (14), GATES (12), DAVIS (7), and Miss DIGBY (8).

The great number of bivalent chromosomes, together with the frequent overlapping of their ends, makes an exact calculation rather difficult. Several counts were made, however, by sketching them with the aid of a camera lucida, and it seems safe to say that there are twenty-four pairs of chromosomes.

Heterotypic mitosis

When the bivalent chromosomes have attained a maximum condensation and contraction, and the nuclear membrane as well as the nucleolus has disappeared, the spindle suddenly appears. The chromosomes then begin to arrange themselves and move toward the equatorial plate. An oblique polar view as the chromosomes move toward the equatorial plate is shown in fig. 2. They exhibit the same difference in size and shape observed in the diakinesis stage. The large chromosome is very conspicuous, with its somewhat wavy outline. The unequal pair is also distinct. In fig. 3, showing a polar view of the plate, about twenty-four bivalent chromosomes can easily be counted. Even in this stage the giant chromosome and the unequal pair are not difficult to distinguish.

A typical side view of the metaphase of the heterotypic mitosis is shown in fig. 4. On account of the large number of chromosomes, all of them cannot be seen distinctly. The giant chromosome as

usual is again distinct in this stage, but the unequal pair is not so easily recognizable. The point of attachment of the spindle does not seem to be definite. Generally it is at the end, but sometimes it is a little toward the middle. The double nature of the daughter chromosomes is practically unrecognizable, but occasionally may be traced more or less from their tips.

An early stage of anaphase, showing the gradual separation of the two members of each pair of chromosomes, is found in fig. 5. The members of one pair have already separated, an early separation characteristic of the unequal pair; another is nearly separated; but the rest, especially the large one, are just commencing to pull apart. A side view of a later stage of the heterotypic figure, in which the two members of the pairs have just separated, is shown in fig. 6. One of the pairs, however, lags somewhat. In this stage the number of chromosomes is fairly easy to determine. The giant chromosome and the small member of the unequal pair are easily distinguished. The two members of the large pair are *V*-shaped, with the apex of the *V* attached to the spindle. A more advanced stage, in which the double nature of the haploid chromosome is more or less distinguishable, is shown in fig. 7. This is usually indicated at their tips, which are clearly parted in most of them into two halves. A side view of a late heterotypic anaphase, in which the daughter chromosomes have just reached the poles, is represented in fig. 8. This is an early stage of telophase, and from this time the chromosomes gradually begin to assemble and pass into an anastomosing condition, which is followed immediately by the formation of the nuclear membrane. The most characteristic feature of the daughter nuclei is that they do not reach the true resting stage characterized by the formation of a reticulum. No satisfactory evidence of a new fission in their limbs could be obtained.

The second meiotic division follows quickly, and therefore the nuclear membrane persists for only a short time before it begins to break down again.

Homotypic mitosis

As the two nuclear membranes break down, the two homotypic spindles appear simultaneously. These may lie parallel or at

right angles to each other. The chromosomes immediately arrange themselves in the equatorial plate and split longitudinally, the line of separation being that already recognized in the anaphase of the preceding division. The daughter chromosomes then move toward the poles.

The fact that in the material there are extremely few stages of the homotypic division, is evidence that as usual they pass through this stage very quickly. A side view of a typical homotypic metaphase is represented in fig. 9, which, however, shows some disarrangement caused by sectioning. In this case the homotypic spindles lie parallel to each other. The chromosomes are as usual of different size and shape. They are more or less rod-shaped, and some are almost globular. The giant chromosome is distinct, while the small one can be seen with difficulty or not at all in this stage. A side view of the four daughter nuclei derived from a single pollen mother cell, which are the nuclei of the four future pollen grains, is shown in fig. 10.

Relation of chromosomes to sex

This brief cytological study of *Elodea gigantea* reveals the fact that the chromosome group in the cells of the male gametophyte contains two large elements which exceed both in length and in thickness any of the other chromosomes, and one element which is smaller than the rest. A typical group, in which the two large chromosomes and the small one are very conspicuous, is shown in fig. 11. On account of the large number of chromosomes and the frequent overlapping of their ends, the number in vegetative division is hard to determine, but several counts indicate that there are about forty-eight. Since the haploid number is twenty-four, it is reasonably certain that forty-eight is the diploid number. The fact that the chromosomes lie at different angles makes it difficult to secure favorable views showing the whole number of the chromosomes, and especially the two giant ones. In most of the figures examined, however, the two giant chromosomes are conspicuous, but the small one is not so easily seen and identified.

In the dividing pollen mother cells already described, a marked difference in the relative size of the chromosomes is also observed.

In the diakinesis stage there are twenty-four bivalent chromosomes, one pair of which is much larger than the others; while another pair is characterized by the unequal length of its component elements. It is evident, therefore, that the two giant chromosomes in the sporophyte have united, and that the small chromosome has paired with a chromosome of normal size, resulting in an uneven pair. In the heterotypic division, one of the daughter nuclei receives one giant chromosome, the larger chromosome of the unequal pair, and twenty-two of regular size; while the other daughter nucleus receives one giant chromosome and a very small one (from the unequal pair), and twenty-two of regular size, to which the conventional term autosome is now being applied. In the homotypic mitosis each of the chromosomes of the daughter nuclei divides equally. Thus two of the resulting nuclei receive one giant and one small chromosome and twenty-two autosomes; while the other two receive one giant chromosome, the larger member of the unequal pair, and twenty-two autosomes. From this it is clear that after the homotypic mitosis two kinds of pollen grains are formed, half of which have a smaller amount of chromatin than the other half.

The presence and behavior of the large and small chromosomes in the somatic and meiotic division in *Elodea* are quite in harmony with the conditions found in certain insects, especially in *Drosophila* (STEVENS **30**, MORGAN **22**, and METZ **20**); *Lygaeus* (WILSON **33**); and *Euschistus* (MONTGOMERY **21**); and also in harmony with the results obtained in *Sphaerocarpus*, a dioecious bryophyte (ALLEN **2**). In view of this similarity, and for the sake of uniformity, the writer will adopt the designations applied to the particular chromosomes in these insects, that is, *X* for the large and *Y* for the small chromosome of the unequal pair. This naming, however, is subject to correction until the presence of the large *X* chromosome is determined definitely from a study of the female plant. The similarity in form and behavior of the *X* and *Y* chromosomes of *Elodea* to those of certain animals is very suggestive. The writer has not the least doubt that the unequal pair of chromosomes bears sex characters as in *Sphaerocarpus* and *Lygaeus*. The function of the large pair of chromosomes still remains to be determined.

Discussion

From this account it is evident that the maturation division in *Elodea* is telosynaptic. It is somewhat in accord with the schemes of Miss DIGBY (**8**) and GATES and REES (**13**). There is an indication that the chromatin is split or apparently double before synizesis. The writer was unable to find a stage where there was not evidence of the double nature. This undoubtedly suggests that the chromatin threads were double from the beginning. This double nature, however, is only revealed with certainty when synizesis is about to take place, at which time the chromatin threads have become very sharply defined. After synizesis the spireme appears to be double. It becomes thickened, and arranges itself into a number of loops corresponding to the gametic number of chromosomes. In this stage it loses its apparent double nature and becomes smooth and single. The loops gradually separate from each other at the base during the segmentation of the spireme. Thus all the chromosomes are formed in the usual way. The segments condense until a rod-shape is acquired.

In the late diakinesis stage the bivalent chromosomes, twenty-four in number, become greatly condensed, but their double nature is still distinguishable, a straight line of fission usually showing between the halves. A difference in size can be seen, the giant chromosome being very conspicuous, while the *Y* (small) chromosome, being attached to the *X* chromosome (of the same size as the autosomes), is not always easy to identify.

As the primary purpose of this paper was not a study of synizesis, the writer has decided to refer only to some recent papers, such as those of Miss DIGBY (**8**), GATES and REES (**13**), and OSAWA (**26**).

In regard to the unequal pair of chromosomes in *Elodea*, all the evidence indicates that they are factors in sex determination. Their rôle in the unequal distribution of chromatin is also very significant. In most of the known cases in animals where there is a difference in the chromosomes, the female has a larger number of chromosomes or a greater quantity of chromatin. WILSON (**34**, **35**), however, believes that these quantitative differences cannot always be considered as the primary sex determining factors; for he found that such view is inapplicable to cases like *Nezara*, *Oncopeltus*, or

Metapodius. In *Nezara* and *Oncopeltus* the idiochromosomes are of equal size, and there is no visible quantitative difference; while on the other hand, in *Metapodius* the male appears to contain more chromatin than the female. In all these genera, however, the phenomena are all otherwise closely similar to those seen in the other insects. In the case of *Elodea*, the writer firmly believes that the unequal pair of chromosomes functions in the determination of sex.

WILSON (**34**), in his comparative review of the types of sexual differences of the chromosome groups, divides the much studied cases into five classes, as follows:

1. Both sexes with same number of chromosomes, a pair of equal idiochromosomes present in both. No visible difference between the two classes of spermatozoa or between the male and female somatic groups.

2. Both sexes and both classes of spermatozoa with the same number of chromosomes, the male with a pair of unequal idiochromosomes, half the spermatozoa receiving the large one and half the small one.

3. The female chromosome group with one or more chromosomes than the male. The male with an unpaired idiochromosome and an odd spermatogonial number, half the spermatozoa receiving the idiochromosome and half being without it.

4. Female group (by inference only) with two more chromosomes than the male. In the male a pair of unequal idiochromosomes, half the spermatozoa receiving both these idiochromosomes, and hence two more than the other half.

5. Female group with three more chromosomes than the male. Half the spermatozoa receiving three more chromosomes than the other half.

It is evident from this classification that despite the apparent diversity of the types, all are in accord with the well known principle that the spermatozoa are of two kinds, equal in number, respectively male producing and female producing. The case of *Elodea* is quite in harmony with those in the second group, and at the same time it is in accord with the principle.

Turning to the question of dioecism, it seems that all the accumulated evidence points very strongly to the conclusion that in dioecious mosses, and perhaps in dioecious Bryophytes in general, the segregation of the sex factors takes place during sporogenesis. The MARCHALS (**16, 17**), in their carefully conducted experiments on mosses, were able to show that two kinds of spores, equal in number,

are produced in the capsule, one of which develops into male gametophytes and the other into female gametophytes. By changing the environmental conditions they were not able to alter the sexes. On the other hand, by means of protonemata regenerated from the tissue of the sporophyte, a leafy branch bearing both antheridia and archegonia was produced. From the experiment it is clear that in the sporophyte both sexes are present, and the segregation of the qualitatively different sex factors takes place in the maturation division, in which two kinds of spores, equal in number, are produced. Similar results were obtained by DOUIN (9), STRASBURGER (32), and ALLEN (2) on *Sphaerocarpus*.

CORRENS (5), working on higher plants, obtained results from his experiments on *Bryonia* hybrids which harmonize remarkably with the cytological behavior of *Elodea*. He came to the conclusion that the egg contains only the female "tendency," and that it is recessive to the male; but two kinds of microspores are formed with both tendencies, the male dominant and the male latent. DARLING (6) came practically to the same conclusion.

In monoecious plants it seems very evident from the experiments that in the mosses the sporophyte is bisexual in character, the two sexes being brought together by fertilization, and that a separation of the sexes occurs during sporogenesis, so that half of the spores produce male gametophytes and half female.

STRASBURGER (32) presents a general discussion of his experiments on *Elodea*, *Mercurialis*, and other plants, a discussion well summarized by SHARP (29), as follows:

> In monoecious mosses the segregation of the sexes takes place in the somatic mitosis at the time the sex organs are formed. The separation has been secondarily joined with reduction, so that in the derived dioecious mosses it occurs at sporogenesis. In the homosporous Pteridophytes it takes place at some stage in the gametophyte before the formation of the sex cells, as in moneocious mosses, although in some cases (*Equisetum*, *Onoclea*, and others) a marked physiological dioecism is present. Heterosporous Pteridophytes may be either monoecious (hermaphroditic) or dioecious. In monoecious forms the sexes are separated at some stage before the development of megaspores and microspores, whereas in dioecious forms it must take place at some other point in the life cycle, since the two kinds of sporophytes (megaspore-bearing and microspore-bearing) are distinct from their initial stages. There is some evidence to show that such dioecism is due to a differentiation among the pollen grains and hence

among the male gametes. Some grains have a strong male tendency which dominates the female tendency of the egg, male progeny resulting; while other grains have a weak male tendency dominated by the female tendency of the egg, female offspring being produced. In brief, as sex separation became joined with reduction in forms with monoecious gametophytes, the dioecism of the gametophyte and heterospory (first physiological and morphological) followed; and this in turn led to dioecism. Finally, in such advanced forms there appears to be a differentiation among the spores of one sex, the microspores giving male gametes of two types.

This quotation evidently agrees very well with the results obtained from the present cytological study on *Elodea gigantea*, that the resulting offspring depends upon the type of male gamete functioning, as is known to be the case in so many animals.

ALLEN suggested that the separation of the sex factors in dioecious seed plants may possibly occur in the division which differentiates the two male nuclei in the pollen tube, rather than at the divisions producing microspores. SHARP believes that this interpretation cannot be applied to Mendelian factors in general, for in maize hybrids the embryo, with very rare exceptions, has been found to be like the endosperm with respect to factors introduced by the pollen parent. With these factors, therefore, the male nuclei appear to be qualitatively similar.

COLLINS' (**3, 4**) results in his experiments on *Funaria hygrometrica*, a monoecious type of moss, are completely in accord with STRASBURGER'S conclusion regarding monoecious mosses. In his experiments he was able to show that the gametophytes arising from spores are all bisexual (monoecious), but the gametophytes produced by regeneration from the antheridia or the surrounding "perigonial" leaves of the "male flower" are male plants only; while the gametophytes derived from the cultures of the venter and neck cells, or the "perichaetial" leaves of the "female flower," are all female plants. From these results COLLINS believes it is possible that the dioecious plants may have been produced as the result of vegetative propagation, following such a somatic division in the tissue of the monoecious gametophyte.

A cytological study of the female plant of *Elodea gigantea*, and a complete study of *E. canadensis* is desirable, but would probably only confirm the conclusions already reached.

Summary

1. The pollen mother cells, before rounding off, have dense, finely granular cytoplasm with a large nucleus.

2. The resting nucleus consists of delicate chromatin threads or meshes, with some nodelike structures at the intersections of the threads.

3. The first indication that reduction is about to take place is the appearance of a clear space at one side of the nucleus, the thickening of the chromatin threads, and the enlargement of the nodes of the chromatin network.

4. During synizesis there is growth of the nucleus and a considerable contraction of the reticulum. The threads become rearranged, and a double continuous filament is formed, which has a granular character.

5. After synizesis the filament, commonly known as the spireme, loosens from a tight knot and extends throughout the nuclear cavity. Beautiful loops are formed, which are gradually detached from the base. The spireme becomes a thickened and smooth filament before the second contraction and segmentation take place.

6. The individual segments unite end to end and gradually condense, sometimes twisting about each other. The method of reduction in *Elodea*, therefore, is telosynaptic.

7. The bivalent chromosomes are twenty-four in number, one pair of which is conspicuously longer and thicker than the others, and another pair is characterized by having unequal members.

8. The chromosomes become shortened and thickened by contraction, while the nuclear membrane becomes thinner and thinner, and finally disappears. This disappearance of the nuclear membrane is followed by the formation of the heterotypic spindle, which draws the chromosomes into the equatorial plate.

9. Each of the bivalent chromosomes is attached to the spindle by one end, and the two constituent chromosomes move apart. The giant (*L*) chromosome splits lengthwise and forms two large daughter chromosomes; while the small (*V*) chromosome separates from the other member of the pair and goes to one pole.

10. As the daughter chromosomes move toward the poles they become *V*-shaped, with the apex of the *V* attached to the spindle. Their double nature is sometimes shown at their tips.

11. At the pole of the heterotypic spindle the daughter chromosomes unite side by side, and cross connections appear.

12. The resulting daughter nuclei from heterotypic division do not pass into a true resting stage, not reaching a true reticulum, but soon reorganizing for the next mitosis. As the nuclear membranes disappear the two homotypic spindles appear simultaneously. The chromosomes arrange themselves in the equatorial plate and then split equally lengthwise.

13. Two of the four resulting nuclei receive one giant (*L*), one small (*Y*) chromosome, and twenty-two of the chromosomes which are now becoming designated as autosomes; while each of the other two receives one giant (*L*) chromosome, the larger member of the unequal pair (*X*), and twenty-two autosomes. Thus, in the reconstruction of the four daughter nuclei, two of them contain more chromatin than the other two. The resulting pollen grains, however, look alike.

14. In the somatic mitosis there are forty-eight chromosomes, two of which are conspicuously longer and thicker than the others, while one is noticeably smaller. In the reduction, the two large chromosomes unite with each other, and the small chromosome unites with one of ordinary size.

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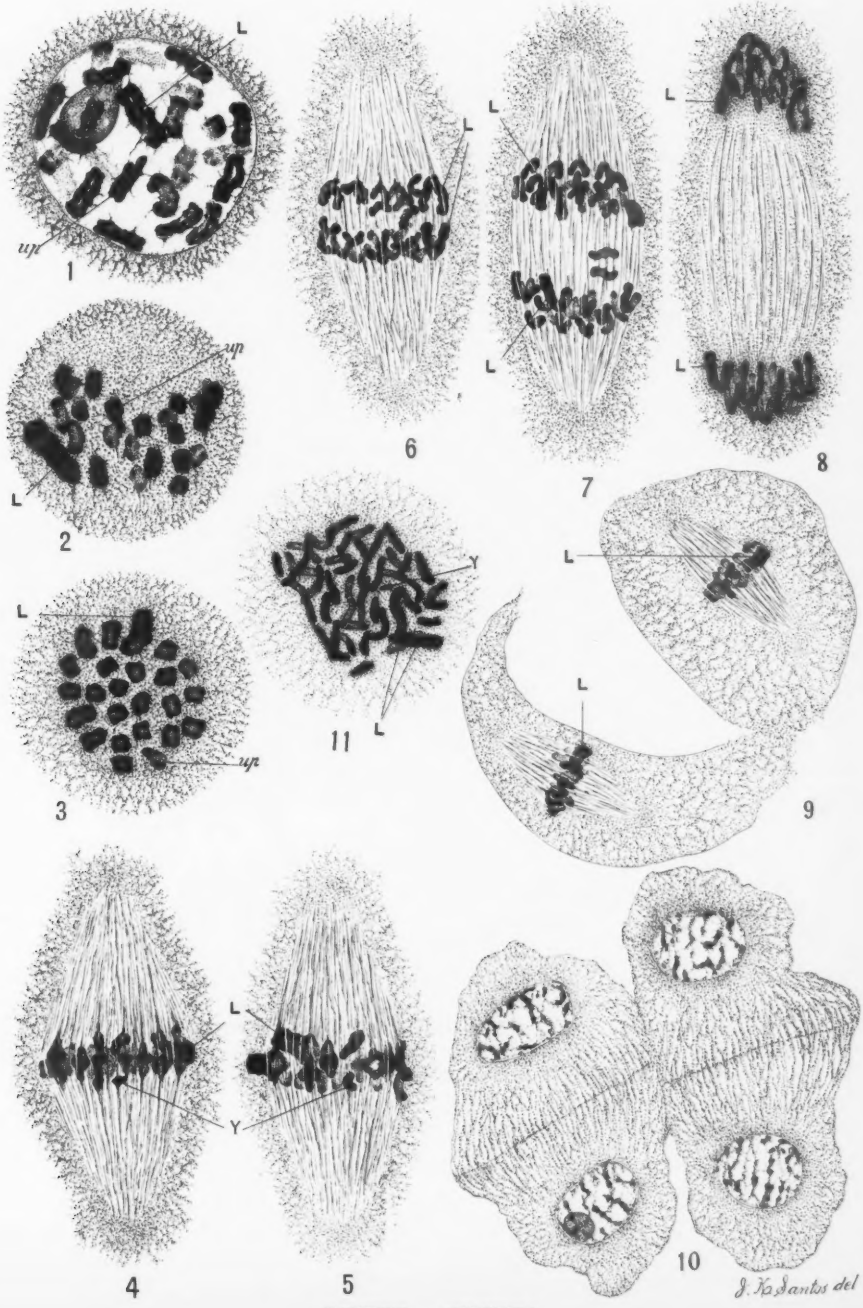
LITERATURE CITED

1. ALLEN, E. C., A chromosome difference correlated with sex differences in *Sphaerocarpus*. Science **46**:466–467. 1917.
2. ———, The basis of sex inheritance in *Sphaerocarpus*. Proc. Amer. Phil. Soc. **58**:289–315. 1919.
3. COLLINS, E. J., Sex segregation in Bryophyta. Jour. Genetics **8**:139–146. 1919.
4. ———, The genetics of sex in *Funaria hygrometrica*. Proc. Roy. Soc. London. Series B. **91**:369–370. 1920.
5. CORRENS, C., Die Bestimung und Vererbung des Geschlechtes. Berlin. 1907.
6. DARLING, C. A., Sex in dioecious plants. Bull. Torr. Bot. Club **36**:177–199. *pls. 12–14*. 1909.
7. DAVIS, B. M., The reduction division of *Oenothera biennis*. Ann. Botany **24**:641–651. 1910.
8. DIGBY, L. On the archesporial and meiotic mitosis of *Osmunda*. Ann. Botany **33**:135–172. *pls. 8–12*. 1919.

9. DOUIN, C., Nouvelles observations sur *Sphaerocarpus*. Revue Bryol. **36**: 37–41. *figs. 10*. 1909.
10. DOUIN, C., and DOUIN, R., Notes sur les *Sphaerocarpus*. Rev. Gen. Bot. **29**: 129–136. 1917.
11. FRASER, H. C. I., The behavior of the chromatin in the meiotic divisions of *Vicia Faba*. Ann. Botany **28**: 633–642. 1914.
12. GATES, R. R., A study of reduction in *Oenothera rubrinervis*. BOT. GAZ. **46**: 1–24. 1908.
13. GATES, R. R., and REES, E. M., A cytological study of pollen development in *Lactuca*. Ann. Botany **35**: 365–398. 1921.
14. HARPER, R. A., Kerntheilung und freie Zellbildung in ascus. Jahrb. Wiss. Bot. **30**: 249–284. 1897.
15. LAWSON, A. A., The phase of the nucleus known as synapsis. Trans. Roy. Soc. Edinburgh **47**: 591–604. 1911.
16. MARCHAL, E., and E., Recherches experimentales sur la sexualité des spores chez les mousses dioiques. Mem. Cour. Acad. Roy. Belg. Cl. Sci. II. **1**: 1. 1906.
17. ———, Aposporie et sexualité chez les mousses. Bull. Acad. Roy. Belg. Cl. Sci. IV. **7**: 765. 1907.
18. MCCLUNG, C. E., The chromosome complex of Orthopteran spermatocytes. Biol. Bull. **9**: 304–339. 1905.
19. ———, The multiple chromosomes of *Hesperotethix* and *Mermiria* (Orthoptera). Jour. Morph. **29**: 519–605. 1917.
20. METZ, C. W., Chromosomes studies in the Diptera. I. A preliminary survey of five different types of chromosome groups in the genus *Drosophila*. Jour. Exp. Zool. **17**: 46–56. 1914.
21. MONTGOMERY, T. H., The spermatogenesis of an Hemipteran, *Euschistus*. Jour. Morph. **22**: 718–731. 1911.
22. MORGAN, T. H., The physical basis of heredity. pp. 305. *figs. 117*. Lippincott. Philadelphia and London. 1919.
23. MOTTIER, D. M., The development of the heterotypic chromosomes in pollen mother cells (*Podophyllum*). Ann. Botany **21**: 309–347. 1907.
24. ———, Mitosis in the pollen mother cells of *Acer Negundo* L. and *Staphylea trifolia* L. Ann. Botany **28**: 115–133. 1914.
25. NOLL, F., Versuche über die Geschlechtsbestimmung bei diocischen Pflanzen. Sitz. Ber. Neiderrhem. Gesells. 1–40. 1907.
26. OSAWA, I., Cytological and experimental studies in *Morus*, with special reference to triploid mutants. Bull. Imp. Sericulture Exp. Sta. Japan. **1**: 318–369. *figs. 8*. 1920.
27. SCHAFFNER, J. H., Dioeciousness in *Thalictrum dasycarpum*. Ohio Jour. Sci. **20**: 25–34. 1920.
28. SHARP, L. W., Somatic chromosomes in *Tradescantia*. Amer. Jour. Bot. **7**: 341–354. 1920.
29. ———, An introduction to cytology. pp. 452. New York: McGraw-Hill Co. 1st ed. 1921.



SANTOS on ELODEA

30. STEVENS, N. M., The chromosomes of *Drosophila ampilophila*. Proc. 7th Internat. Zool. Congress, Boston. 1907.
31. STRASBURGER, E., Versuche mit diocischen Pflanzen in Riicksicht auf Geschlechtsverteilung. Biol. Centralbl. **20**: 657. 1900.
32. ———, Über geschlechtbestimunde Wischen. Jahrb. Wiss. Bot. **48**: 427–520. 1910.
33. WILSON, E. B., Studies on chromosomes. I. The behavior of the idiochromosomes in Hemiptera. Jour. Exp. Zool. **2**: 371–406. 1905.
34. ———, Studies on chromosomes. IV. The "accessory" chromosomes in *Syromastes* and *Pyrrochoris*, with a comparative review of the types of sexual differences of the chromosome groups. Jour. Exp. Zool. **6**: 68–99. 1909.
35. ———, Studies on chromosomes. V. The chromosomes of *Metapodius*. A contribution to the hypothesis of the genetic continuity of chromosomes. *Ibid.* **6**: 147–205. 1909.
36. ———, The sex chromosomes. Archiv. Fur. Mikroskopische Anatomie **77**: 249–268. 1911.

EXPLANATION OF PLATE III

FIG. 1.—Median section of nucleus at diakinesis stage, with about twenty-four bivalent chromosomes which seem to have attained their maximum size; *L*, giant chromosome, *UP*, unequal pair.

FIG. 2.—Oblique polar view as chromosomes move toward equatorial plate.

FIG. 3.—Polar view of plate, in which twenty-four bivalent chromosomes appear more or less rod-shaped and cubical.

FIGS. 4, 5.—Two typical views of heterotypic metaphase, showing actual splitting of chromosomes; *Y*, small chromosome from unequal pair.

FIG. 6.—Side view of anaphase, in which two members of the pairs have just separated; most of them appear *V*-shaped, with apex of *V* attached to spindle.

FIG. 7.—More advanced stage of heterotypic anaphase; *Y* chromosome not easily distinguished, while *L* chromosomes conspicuous..

FIG. 8.—Early stage of telophase; daughter chromosomes just reached poles and becoming somewhat elongated; *L* chromosomes distinct, *Y* chromosome indistinguishable.

FIG. 9.—Side view of homotypic mitosis; daughter cells somewhat split apart, caused by sectioning; in this case spindles lie parallel to each other; *L* giant daughter chromosomes ready to divide.

FIG. 10.—Side view of four daughter nuclei derived from single nucleus of pollen mother cell.

FIG. 11.—Typical polar view of group of somatic chromosomes; *L*, two monivalent chromosomes; *Y*, small chromosome, one of members of the unequal pair.

FLOWERS AND INSECTS. XXII

CHARLES ROBERTSON

Pollination

Allogamy (Cross-fertilisation, DARWIN; Dicogamia, DELPINO[1] **10,** 4; Fremdbestäubung, MÜLLER **41,** 111, mistranslated "cross-fertilisation" in **42,** 69; Allogamie, in part, Xenogamie, KERNER **25,** 192); cross-pollination: pollination takes place between flowers of different plants.

Gitonogamy (Fremdbestäubung, MÜLLER **41,** 212, **42,** 223; Self-fertilisation, DARWIN **8,** 132, 329; Allogamie, in part, Geitonogamie, KERNER **25,** 192; Gitonogamie, E. and G. **18,** 68): pollination between flowers of the same plant.

Forms of flowers with reference to pollination

The following arrangement is based on that of ERRERA and GEVAERT (**18**). The first references often refer to the condition, the latter to the designation. In the literature the designation is credited sometimes to the author who discovered the condition, sometimes to the one who suggested the designation, and sometimes to the one who originated the designation.

FLOWERS PERFECT (ERRERA and GEVAERT **18,** 162)

Dichogamy (Dichogamia, Dichogamie, SPRENGEL **46,** 19; Asincronogonismo, DELPINO **16,** 156): opposed, Homogamy (Homogamie, SPRENGEL **46,** 19; Singinandria, DELPINO **14,** 156): the anthers and stigmas are mature at different times.

Proterandry (PONTEDERA **43,** SPRENGEL **46,** 158; LINNAEUS; AXELL **1,** 3, 33; KÖLREUTER **30,** 34; SPRENGEL **46,** 227; Dichogamia androgyna, männlich-weiblich Dichogamie, SPRENGEL **46,** 19; Protandrische Dichogamen, HILDE-

[1] See *American Naturalist* **3**: 42. 1876, for a review of DELPINO, in which dicogamia is translated dichogamy instead of cross-fertilization, and called a "modest" title. DAVIS (**29,** 14), instead of transcribing the title of DELPINO'S work, as KNUTH does, mistranslates it in the text and gives the original title in a footnote.

BRAND **23,** 17; Proterandria, DELPINO **11,** 10): the anthers discharge their pollen before the stigmas are receptive.

Proterogyny (Dichogamia gynandra, weiblich-männlich Dichogamie, SPRENGEL **46,** 19; Protogynische Dichogamen, HILDEBRAND **23,** 18; Proteroginia, DELPINO[1] **11,** 10): the stigmas are receptive before the anthers discharge.

Hercogamy (SPRENGEL; Flores hercogami, Herkogami, AXELL **1,** 40, 86; Ercogamia, DELPINO **14,** 183): spontaneous self-pollination is prevented by relative positions of anthers and stigmas.

Adynamandry (KÖLREUTER **30**; DARWIN **8,** 330; Adinamandria, DELPINO **14,** 200): self-sterility: pollen from the same flower fails to effect fertilization.

Prepotency (DARWIN **8,** 391): pollen from another individual is prepotent over the plant's own pollen.

Xenautogamy: flowers homogamous, or nearly so, favoring cross-pollination, but regularly self-fertile under adverse conditions.

Autogamy (VAILLANT **47**; LINNAEUS; Spontaneous self-fertilisation, DARWIN; Autogamia, DELPINO **10,** 36; Omogamia, DELPINO **12,** 25); self-pollination: pollination occurs between the anthers and stigmas of the same flower.

Direct autogamy (VAILLANT **47**; LINNAEUS; ERRERA and GEVAERT): spontaneous self-pollination: occurs as the result of movements or changes in the flower.

Indirect autogamy (LINNAEUS; ERRERA and GEVAERT): is effected by some pollinating agency.

Pseudoclistogamy (Pseudokleistogame Blüten, HANSGIRG **19**; Pseudo-Kleistogamie, KNUTH **28,** 35): the flowers, which normally open, remain closed under certain conditions.

Photoclistogamy (Photokleistogame Blüten, HANSGIRG **19**; Photokleistogamie, KNUTH **28,** 35): on account of a deficiency of light.

[1] The use of DELPINO's terms proterandry and proterogyny seems to be connected with the fact that HILDEBRAND never used the nouns protandry and protogyny.

Hydroclistogamy (HILDEBRAND **23,** 77; Hydrokleistogame Blüten, HANSGIRG **19;** Hydrokleistogamie, KNUTH **28,** 35): on account of being submerged.

Thermoclistogamy (LINNAEUS, BENNETT, DARWIN **9,** 342; Thermokleistogame Blüten, HANSGIRG **20,** Thermokleistogamie, KNUTH **28,** 35): on account of a deficiency of heat.

Xeroclistogamy (TORREY and GRAY, DARWIN **9,** 330; Xerokleistogame Blüten, HANSGIRG **20**): on account of dryness.

Ombroclistogamy (KERNER **26,** 390): on account of rainy weather.

Clistogamy (DILLENIUS **17;** LUBBOCK **36a,** 33; Cleistogame, KUHN **31,** 66; Kleistogami, AXELL **1,** 78; Cleistogamia, DELPINO **16,** 148): opposed, Chasmogamy (Flores chasmogami, Kasmogam blomma, AXELL **1,** 14; Chasmogamie, E. and G. **18,** 162): the flowers remain closed.

FLOWERS PLEOMORPHOUS (ERRERA and GEVAERT **18,** 162)

Chasmoclistogamy (Dimorphe Blüten, VON MOHL **38;** Blütendimorphismus, HILDEBRAND **23,** 73; Blütenpolymorphismus, KUHN **31,** 65–67; Individui casmocleistogamici, DELPINO **16,** 151; Chasmo-cleistogamie, E. and G. **18,** 162): opposed, Archoclistogamy (BURCK **3,** 17–20; KNUTH **29,** 57; Archokleistogamie, KNUTH **28,** 71; archocleistogamous flowers, **29,** 58): some of the flowers are chasmogamic, the others clistogamic.

Agamonecism (SPRENGEL **46,** 33; Agamo-monoecie, E. and G. **18,** 162): the flowers are perfect and neutral.

Andromonecism (LINNAEUS **34,** 93; Plants andro-monoecious, DARWIN **9,** 12; Andro-monoecie, E. and G. **18,** 145): flowers staminate and perfect.

Gynomonecism (LINNAEUS **34,** 93; Plants gyno-monoecious, DARWIN **9,** 12; Gyno-monoecie, E. and G. **18,** 145): flowers pistillate and perfect.

Trimonecism (LINNAEUS; Monoicous polygamous plants, DARWIN **9,** 12; Tri-monoecie, E. and G. **18,** 145, 162; Cenomonoecie, KIRCHNER **27**): flowers staminate, pistillate, and perfect.

Monecism (Monoecia, LINNAEUS; Monoeci, AXELL **1**, 95): flowers staminate and pistillate.

Ecodichogamy (Dichogamy, authors): monecious asynchronism.

Ecoproterandry (MARTIUS **37**; DELPINO **14**, 306): the staminate flowers mature before the pistillate.

Ecoproterogyny (DELPINO **12**, 18, **14**, 158): the pistillate flowers mature before the staminate.

PLANTS PLEOMORPHOUS (ERRERA and GEVAERT).

Diëcodichogamy (Dimorphismo nel tempo, DELPINO **15**, 338, 342): some individuals ecoproterandrous, the others ecoproterogynous.

Heterostyly (Heterostylie, HILDEBRAND **23**, 33, 46): opposed, Homostyly (Nichtheterostylie, HILDEBRAND **23**, III; Homostyli, AXELL **1**, 95): the flowers have different forms depending upon the relative lengths of stamens and styles.

Heterodistyly (SPRENGEL **46**, 103; Dimorphism, DARWIN **5**; Heterostyled dimorphic plants, DARWIN **9**, 14; Heterodistylie, E. and G. **18**, 163): one form with short stamens and usually a long style, another with long stamens and usually a short style.

Heterotristyly (VAUCHER **48**, **2**:371; DARWIN **9**, 138; Trimorphism, DARWIN **6**; Heterostyled trimorphic plants, DARWIN **9**, 137; Heterotristylie, E. and G. **18**, 163): a long-styled form with short and mid-length stamens, a mid-styled form with short and long stamens, a short-styled form with mid-length and long stamens.

Autallogamy (MÜLLER **42**, 118; Omodicogamia, DELPINO **16**, 142; Auto-allogamie, E. and G. **18**, 147, 163): one individual with flowers autogamic, another with flowers allogamic.

Homodichogamy (Homo-dichogamie, E. and G. **18**, 163): some individuals with flowers homogamous, others with flowers dichogamous.

Heterodichogamy (MÜLLER **42**, 441; Heterodichogamie, E. and G. **18**, 152, 163, in part): some individuals with perfect flowers proterandrous, others with perfect flowers proterogynous.

Hometerostyly (WARMING **49,** 114; Homo-Heterostylie, KNUTH **28,** 61): some individuals with flowers homostyled, others with flowers heterostyled.

Androdiecism (LINNAEUS **34,** 95; Plants andro-dioecious, DARWIN **9,** 13; Andro-dioecie, E. and G. **18,** 153, 163): one individual with flowers staminate, another with flowers perfect.

Gynodiecism (SPRENGEL **46,** 310; Plants gyno-dioecious, DARWIN **9,** 12; Gyno-dioecie, E. and G. **18,** 153, 163): one individual with flowers pistillate, another with flowers perfect.

Triecism (LINNAEUS **34,** 95; Trioicous polygamous plants, DARWIN **9,** 11; Trioecie, E. and G. **18,** 159, 163); one individual with flowers staminate, another with flowers pistillate, another with flowers perfect.

Diecism (Dioecia, LINNAEUS; Dioeci, AXELL, **1,** 95): one individual with flowers staminate, another with flowers pistillate.

Phytodichogamy (Dichogamy, authors): diecious asynchronism.

Phytoproterandry: the staminate individuals begin to bloom before the pistillate. No examples (?).

Phytoproterogyny (KERNER **26,** 313): the pistillate individuals begin to bloom before the staminate.

Pleogamy (Pleogamia, LEOW **35,** 197): includes three forms.

Andropleogamy (SCHULZ **44, 45;** Männlich Pleogamie, LOEW **36,** 377): one individual with flowers staminate, another with flowers perfect, another andromonecious.

Gynopleogamy (SCHULZ **44, 45;** Weiblich Pleogamie, LOEW **36,** 377): one individual with flowers pistillate, another with flowers perfect, another gynomonecious.

Eripleogamy (SCHULZ **44, 45;** Pleogamie, Dritte Gruppe, LOEW **36,** 377): one individual with flowers staminate, another with flowers pistillate, another with flowers perfect, the fourth andromonecious, the fifth gynomonecious.

AUTOGAMY.—Except for plants depending upon annual seed-production, the importance of autogamy is generally exaggerated. In the following list cases resulting from direct contact and from pollen fall are arranged chronologically in two sets. Pollen fall,

a more uncertain mode of autogamy, is more obvious in an erect flower where the fall is toward the stigma than in a pendulous flower where the fall is by the stigma. When all of the pollen remains in a flower, a great variety of conditions may favor its touching the stigma, but the probability of effective autogamy is rather discredited, when it depends upon pollen fall and the concurrence of several other precarious conditions. For KERNER'S categories of autogamy (1–20), see KNUTH **29,** 34–36. KERNER does not cite any authors for the particular cases. According to AXELL (**1,** 2; MÜLLER **42,** 27), VAILLANT (**47**) first observed autogamy by movements of stamens and pistils.

1. Filaments bend so that anthers touch stigma (KERNER 6, 7).—*Saxifraga tridactylites*, LINNAEUS **32**; SPRENGEL **46,** 245; KERNER **26,** 337.—*Agrimonia eupatoria*, MÜLLER **42,** 235.

2. Inflection or bending of style (KERNER 10, 11).—*Nigella arvensis*, WAHLBOM **33.**—*Lilium martagon*, SPRENGEL **46,** 188.

3. Bending of style branches.—*Passiflora*, LINNAEUS; Medicus; SPRENGEL **46,** 163.—*Malva rotundifolia*, MÜLLER **42,** 143.

4. Pendulous pollinia and wind.—*Ophrys apifera*, BROWN **2,** 740; DARWIN **4,** 53, 54.

5. Inflection of stigma (KERNER 12).—*Salvia hirsuta*, HILDEBRAND **22.**

6. Anthers touch stigma and on opening pollinate it (KERNER 1).—*Oxalis sensativa*, HILDEBRAND **23,** 71.

7. Fall of tubular corolla with anthers below stigmas (KERNER 15).—*Calceolaria pinnata*, HILDEBRAND **24**; MÜLLER **42,** 431.—*Anchusa officinalis*, MÜLLER **42,** 411.

8. Pollen emits tubes while still in anthers.—*Epipactis viridiflora*, MÜLLER **39**; DARWIN **4,** 102.

9. Flower only partially opening or remaining closed.—*Chelidoniummajus*, MÜLLER **42,** 94.

10. Lengthening of pistil (KERNER 9).—*Stachys betonica*, MÜLLER, **42,** 488.

11. Bending inward of anther.—*Berberis vulgaris*, MÜLLER **42,** 92.—*Brassica oleracea*, MÜLLER **42,** 112.

12. Closing of flower (KERNER 15).—*Lepidium sativum*, MÜLLER **42,** 111.

13. Withering of flower.—*Veronica officinalis*, MÜLLER **42**, 441.

14. Lengthening of stamens and closing of flower.—*Geum rivale*, MULLER **42**, 229.

15. Elongation of corolla with adnate stamens (KERNER 15).—*Myosotis versicolor*, MÜLLER **40**; HENSLOW **21**, 375.

16. Corolla and stamens lengthen.—*Odontites serotina*, MÜLLER **42**, 446.

17. Elongation of receptacle.—*Myosurus minimus*, MÜLLER **40**, **42**, 73.

18. Stamens lengthen (KERNER 5).—*Ipomoea purpurea*, DARWIN **8**, 28.

19. Crossing of style branches.—*Aster alpinus*, KERNER **26**, 359.

20. Shortening of style (KERNER 8).—*Cereus dasyacanthus*, KERNER **26**, 347.

21. Spiral coiling of filaments and styles (KERNER 13).—*Portulaca oleracea*, KERNER **26**, 358.

POLLEN FALL

22. Orchids.—*Neotinea intacta*, MOGGRIDGE; DARWIN **7**, **4**, 27.

23. Lateral flowers.—*Verbascum nigrum*, MÜLLER **42**, 430.

24. Erect flowers (KERNER 4).—*Syringa vulgaris*, MÜLLER **42**, 393.

25. Pendulous flowers (KERNER 2).—*Galanthus nivalis*, MÜLLER **42**, 560.

26. Style bends to line of pollen fall.—*Rhinanthus cristagalli*, *R. minor*, VAUCHER **48**, **3**:539; DELPINO **13**, 133; MÜLLER **42**, 455.—*Solanum tuberosum*, MÜLLER **42**, 426.

27. Recurving stigma touches hairs upon which pollen has fallen.—*Ballota nigra*, MÜLLER **42**, 499.

28. Inflection of stamens.—*Ligustrum vulgare*, MÜLLER **42**, 394.

29. Inflection of flower stalk (KERNER 16).—*Fragaria vesca*, MÜLLER **42**, 231.

30. Flower closing and nodding.—*Helianthemum vulgare*, MÜLLER **42**, 117.

31. Closing of flower dusted with pollen.—*Ranunculus*, HENSLOW **21**, 348.

32. Inflection of corolla (KERNER 15).—*Pedicularis incarnata*, KERNER **26**, 376.

33. Inflection of flower stalk and elongation of sepals (not petals) (KERNER 19).—*Pulsatilla vernalis*, KERNER **26,** 389.

34. Combination of movements of flower stalks, stamens, and style (KERNER 17).—*Ornithogalum nutans*, KERNER **26,** 380.

35. Inflection of flower stalks and styles with elongation of petals and stamens (KERNER 20).—*Geum rivale*, KERNER **26,** 390 (see no. 14).

HOMOSTYLY.—In a table showing HILDEBRAND'S classification, MÜLLER (**42,** 12; KNUTH **28,** 14, 34; **29,** 11, 29) writes homostyly in parenthesis, but that word does not occur in HILDEBRAND'S work.

GITONOGAMY.—This may result from some pollinating agency or spontaneously. MÜLLER observed gitonogamy by the stigma touching the anthers of neighboring flowers, and by pollen fall. Autogamy was observed in three out of four of these cases. Of the cases mentioned by KERNER, which he regards as adaptations for gitonogamy, several were previously described by him as autogamous. Gitonogamy will hardly account for the origin of crowded inflorescences, since these facilitate visits of insects of all kinds, except Sphingidae, and are quite common where spontaneous gitonogamy is impossible, as in galeate Labiatae, Papilionaceae, and Asclepiadaceae.

DICHOGAMY.—In some flowers the dichogamy is complete, but usually there is a homogamous stage. Often the dichogamy is so slight that a flower is called dichogamous by one writer and homogamous by another. Homodichogamous species have dichogamous and homogamous individuals. Heterodichogamous species have proterandrous and proterogynous individuals. In order to distinguish dichogamous flowers, dichogamous individuals, and dichogamous species, monecious dichogamy is designated as *ecodichogamy* and diecious dichogamy as *phytodichogamy*. The latter may be only a difference common in individuals of the same species.

ALLOGAMY.—Allogamy is likely in individuals having single flowers, or in those having a few flowers which are visited in a particular order. It is fairly certain in ecodichogamous plants, and is the only form of pollination possible in individuals having only pistillate flowers, or all of whose flowers are mutually adynamandrous.

CLISTOGAMY.—Some forms of clistogamy seem to be persistent forms of photoclistogamy. KERNER (**26,** 395) observed that *Viola sepincola* does not form open flowers in shady woods, but forms them in open places. *V. cucullata* produces open flowers before the woods become shady and again after the leaves fall, but produces clistogamic flowers in the interval. A patch of *V. striata* which I had under observation produced open flowers until the woods became shady, but plants growing in the thicket margin produced as many open flowers as usual after the shaded plants had ceased.

Lamium amplexicaule (KERNER **26,** 395) behaves in an opposite manner to *Viola cucullata*, for it produces clistogamic flowers in the spring and fall and open ones in summer. This has an appearance of thermoclistogamy. *Amphicarpa* produces clistogamic flowers whose fruits are buried in the ground. This suggests photoclistogamy, but *A. pitcheri* also produces clistogamic flowers at the tips of the aerial branches.

The origin of clistogamic flowers has been supposed to be connected with a deficiency of insects, which may be true in some cases, but all clistogamous flowers are not entomophilous. Clistogamy enables plants to prolong their blooming seasons into unfavorable parts of the year, to produce seeds in localities unfavorable for plants with open flowers, and on parts of the plant which are hidden from the sunlight.

PSEUDOCLISTOGAMY.—This term may be used as a general term to include the different forms of false clistogamy, and in a more special sense to include cases where the conditions are mixed.

HYDROCLISTOGAMY.—The validity of hydroclistogamy as a category is doubtful. It is questionable whether otherwise normal flowers will remain closed only on account of submersion. At Inverness, Florida, I have seen flowers of *Utricularia inflata*, in clear water, open in sunlight, although submerged three feet below the surface (**9,** 311, footnote). Hydroclistogamy is probably associated with submersion in cold, muddy water. The cases mentioned by investigators should probably be called ombroclistogamic.

THERMOCLISTOGAMY.—In cases of flowers called thermoclistogamic, it is doubtful whether cold is the exclusive condition. These should probably be called ombroclistogamic.

OMBROCLISTOGAMY.—This term is proposed to include cases involving the mixed conditions occurring in stormy or rainy weather in which there is an association of cold, excess of moisture, and absence of sunlight.

PHOTOCLISTOGAMY.—Photoclistogamy is not a perfectly satisfactory category, because absence of sunlight is often associated with other unfavorable conditions. It must be remembered here that some flowers open only at night, so that photoclistogamy only affects plants which ordinarily open in sunlight. Xeroclistogamy and ombroclistogamy seem to be the most satisfactory categories, and this might not be so but for the fact that ombroclistogamy includes more than one factor, and xeroclistogamy seems to be associated with conditions affecting the whole plant, not the flowers exclusively.

PARTHENOGENESIS.—Most of the parthenogenetic plants are diecious. Some are entomophilous and some are anemophilous. It seems to be an important condition of seed production when cross-pollination fails. It is not easy to see any advantage when it occurs in perfect flowers which are both allogamic and autogamic. *Populus* may be parthenogenetic. It seems to have too many seeds for an anemophilous plant.

PRIMITIVE SPERMATOPHYTES.—It seems probable that these were perennial plants in which annual seed production was not a condition of natural selection.

ALLOGAMY AND EVOLUTION.—MÜLLER (**42**, 593) and others held that the primitive flowers were diclinic and anemophilous. If so, all flowers were developed from forms in which self-pollination was impossible, so that it was not necessary to explain how flowers came to be cross-pollinated, but how they came to be perfect and some finally clistogamic. If allogamy was an important condition of selection, how did plants which could only be crossed change to a condition in which autogamy is possible or inevitable? How were so many forms of allogamy developed, when the original form showed the most definite of allogamic conditions? The view held here is that the original flowers were perfect and entomophilous, and had the common allogamic characters. After all of the changes and the multiplication of species, only a few plants have flowers which cannot be cross-pollinated. The original flowers and

the forms developed from them are cross-pollinated, so that in the particular cases one does not have to account for allogamy. The theory of the advantage of cross-fertilization may explain the general rule.

ADAPTATION.—Under the theory of natural selection there has been a tendency to regard all floral structures as adaptations, and to assume that they were useful to the plant. From this standpoint adaptation means a condition of selection. It is not always easy to determine whether structures are adaptive or not. To interpret adaptations correctly it is necessary to know the changes of form which have occurred in the past and the changes of conditions through which the plant has gone. No one has such knowledge. Too ready assertion or denial of adaptive significance, therefore, is altogether presumptuous. No one has thorough knowledge of the present conditions which surround plants. The literature contains many statements which show want of discrimination between the natural surroundings of plants and conditions which are the result of human disturbance. No doubt many plant structures have had the effect of actual conditions of selection. It seems evident that, when plants have been selected on account of some decisive advantage, they should be expected to show a rather high degree of perfection in parts only indirectly associated with the condition of selection.

Commenting on KERNER'S so-called arrangements for protection of pollen, KNUTH (**29**, 81) says: "In many plants several of these protective devices are present. Most of the arrangements for the protection of pollen are otherwise advantageous, especially with regard to the possibility of self-fertilization, and the protection of nectar." In this case it is necessary to distinguish arrangements in which the protection of pollen is the only result. Even these may be rejected, if they can be shown to have involved no condition of selection. All of the rest are mere incidental effects, or accidental benefits, and if mentioned at all, should be distinguished as such. The same discrimination should be applied to all so-called adaptations, contrivances, and arrangements.

DARWIN, at least by implication, has disposed of most of the floral modifications so far as concerns their interpretation as adaptations for allogamy. For example, regarding dioecism he

states: "As we must assume that cross-fertilisation was assured before an hermaphrodite could be changed to a dioecious plant, we may conclude that the conversion has not been effected for the sake of gaining the great benefits of cross-fertilisation" (9, 279). Regarding self-sterility he says: "Nevertheless, the belief that self-sterility is a quality which has been gradually acquired for the special purpose of preventing self-fertilisation must, I believe, be rejected" (8, 345).

NATURAL SELECTION AND EVOLUTION OF ANGIOSPERMS.—Probably the most important condition was the diversification of station and habitat. Whenever the ordinary places were filled, it would be an advantage for some plants to change so as to take the unoccupied situations. This diversification would result in plants occupying positions, or migrating to regions, unfavorable for insect visits. In absence of insects perfect flowers may resort to anemophily or autogamy, and diclinic ones to anemophily, hydrophily, or even to parthenogenesis. The dominant perennials occupy the more favorable situations and crowd out or overshadow the other plants. There are places unfavorable for perennials, however, such as bare patches caused by the plow, by tramping of animals, by erosion, by fire, and places under water part of the year. These temporary situations are favorable for annuals. Some annuals are in no respect inferior to perennials as regards their ability to avail themselves of the services of insects, but some situations are only favorable provided the plant can complete its vegetative cycle in a short time. In a general way this means that the plant must be small, so as to develop bloom, and scatter its seeds in a short time. No doubt natural selection often operates in favor of those which can do this the soonest. Here some kind of pollination is necessary and autogamy is general. In the spring many annuals bloom and scatter their seeds before they are overshadowed by more vigorous perennials. Before plowing in the spring the fields are occupied by a considerable flora of this kind. These flowers are as well suited for the visits of bees and other insects as are the larger flowers, but they cannot dispense with the power of autogamy on account of their short seasons and the necessity of annual seed production.

DIVERSIFICATION OF POLLINATING AGENCIES.—In anthecology the separation of flowers into groups is an interesting study, but as regards natural selection it is far more important to show how related plants have become diversified so as to make use of different pollinating agencies. Flowers may be arranged into more than thirty groups, depending upon various means of pollination. The local monocotyledons fall into nine classes, the Ranales into six, and the Polemoniales into five. *Lobelia* has forms pollinated by birds, bumblebees, and other long tongued and short tongued bees.

CARLINVILLE, ILL.

LITERATURE CITED

1. AXELL, S., Om anordningarna for fanerogama växternas befruktning. 1869.
2. BROWN, R., On the organs and mode of fecundation in Orchideae and Asclepiadeae. Trans. Linn. Soc. **16**:683–746. 1833.
3. BURCK, W., Ann. Jard. Bot. Buitenzorg **4**:17–20. 1883.
4. DARWIN, C. R., Fertilisation of orchids. 1862. 2d ed. 1877.
5. ———, On the two forms, or dimorphic condition, in the species of *Primula* and on their remarkable sexual relations. Jour. Linn. Soc. Bot. **6**:77–96. 1862.
6. ———, On the sexual relations of the three forms of *Lythrum salicaria*. Jour. Linn. Soc. Bot. **8**:169–196. 1864.
7. ———, Notes on the fertilisation of orchids. Ann. Mag. Nat. Hist. IV. **4**:141–158. 1869.
8. ———, The effects of cross and self-fertilisation in the vegetable kingdom. 1876.
9. ———, The different forms of flowers on plants of the same species. 1877. 2d ed. 1880.
10. DELPINO, F., Sugli apparecchi della fecondazione nelle piante antocarpee. 1867.
11. ———, Sull'opera "La distribuzione dei sessi nelle piante" del Prof. F. HILDEBRAND. 1867.
12. ———, Ulteriori osservazioni sulla dicogamia nel regno vegetale. Atti Soc. Ital. Sci. Nat. **11**:265–332. 1868. (**1**:1–68).
13. ———, Ulteriori osservazioni Atti **12**:21–141; 179–233. 1869. (**1**:69–189).
13a. ———, Ulteriori osservazioni Atti **13**: 1870. (2^1:1–41).
14. ———, Ulteriori osservazioni Atti **16**:151–349. 1873. (2^2:1–201. 1875).
15. ———, Ulteriori osservazioni Atti **17**: 1874. (2^2:201–351. 1875).
16. ———, Dicogamia e omogamia nelle piante. Nuovo Giorn. Bot. Ital. **8**:140–161. 1876.
17. DILLENIUS, Hortus Elthamensis **2**:329. 1732.

18. ERRERA, L., and GEVAERT, G., Sur la structure et les modes de fecondation des fleurs. Bull. Soc. Bot. Belg. **17**:38–248. 1878.
19. HANSGIRG, A., Phytodynamische Untersuchungen. Bot. Centralb. **41**:397. 1889.
20. ———, Physiologische und phykophytologische Untersuchungen. Bot. Jahrb. **21**:346. 1893.
21. HENSLOW, G., On the self-fertilisation of plants. Trans. Linn. Soc. II. **2**:317–398. 1880.
22. HILDEBRAND, F., Über die Befruchtung der *Salvia*-Arten mit Hülfe von Insekten. Jahrb. Wiss. Bot. **4**:451–477. 1866.
23. ———, Die Geschlechter-Verteilung bei den Pflanzen. 1867.
24. ———, FEDERIGO DELPINO's Beobachtungen über die Bestäubungsvorrichtungen bei den Phanerogamen. Bot. Zeit. **25**:265, 273, 281. 1867.
25. KERNER VON MARILAUN, A., Die Schutzmittel der Blüten gegen unberufene Gäste. Festschr. Zool. Bot. Gesells. Wien. 1876.
26. ———, The natural history of plants. II. 1894. Transl. of Pflanzenleben. 1891.
27. KIRCHNER, O., Flora von Stuttgart und Umgebung. 1888.
28. KNUTH, P., Handbuch der Blütenbiologie. **1**:1–400. 1898.
29. ———, Handbook of flower pollination. **1**:1–382. 1906. Transl. of **28**.
30. KÖLREUTER, D. J. G., Vorläufige Nachricht von einigen das Geschlecht der Pflanzen betreffenden Versuchen und Beobachtungen. 1761. Fortsetzungen 1, 2, 3. 1763, 1764, 1766.
31. KUHN, M., Einige Bemerkungen über *Vandellia* und den Blütenpolymorphismus. Bot. Zeit. **25**:65–67. 1867.
32. LINNÉ, C. VON, Flora Suecica. Ed. 2. 98. 1745.
33. ———, (WAHLBOM, J. G.) Sponsalia Plantarum. 1746.
34. ———, Phil. Bot. Ed. 2. 1780.
35. LOEW, E., Die Veränderlichkeit der Bestäubungseinrichtungen bei Pflanzen derselben Art. Humboldt. **8**:178–183, 214–218. 1889.
36. ———, Blütenbiologische Floristik des mittleren und nördlichen Europa sowie Grönlands. 1894.
36a. LUBBOCK, J., On British wild flowers considered in relation to insects. 1875.
37. MARTIUS, Hist. Gener. Palmarum. **1**:143.
38. MOHL, H. VON, Einige Beobachtungen über dimorphe Blüten. Bot. Zeit. **21**:309–315; 321–328. 1863.
39. MÜLLER, H., Beobachtungen an westfälischen Orchideen. Verh. Natur. Vereins **25**:1–62. 1868.
40. ———, On the fertilisation of flowers by insects and on the reciprocal adaptations of both. Nature **10**:129–130. 1873.
41. ———, Die Befruchtung der Blumen durch Insekten und die gegenseitig Anpassungen beider. 1873.
42. ———, The fertilisation of flowers. 1883. Transl. of **41**.
43. PONTEDERA, G., Anthologia, seu de floris natura. 1720.

44. SCHULZ, A., Beiträge zur Kenntniss der Bestäubungseinrichtungen und Geschlechtsverteilung bei den Pflanzen. I. 1888.

45. ———, *Ibid.* II. 1890.

46. SPRENGEL, C. K., Das entdeckte Geheimniss der Natur im Bau und in der Befruchtung der Blumen. 1793.

47. VAILLANT, S., Discours sur la structure des fleurs, leurs differences et l'usage de leurs parties. 1717.

48. VAUCHER, S. P. E., Histoire physiologique des plantes d'Europe. Vols. I–IV. 1841.

49. WARMING, E., Om bygningen og den formodede Bestövningsmaade af nogle grönlandske Blomster. Vet. Ak. Overs. Kjöbenhavn. 101–159. 1886.

DEVELOPMENT OF THE PROTHALLIUM OF LYGODIUM PALMATUM

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(WITH PLATES IV–VI)

Several earlier workers have reported on the prothallia of the Schizaeaceae. KNY (10) studied the prothallia of *Aneimia hirta;* BURCK (5) the prothallia of *A. Phyllitidis, A. fraxinifolia,* and *A. longifolia;* and BAUKE (2) those of *A. Phyllitidis, A. cheilanthoides, A. collina, Mohria Caffrorum,* and (3, 4) of *Lygodium japonicum.* Later, HEIM (9) studied those of *Aneimia Dregeana, A. fraxinifolia, A. Phyllitidis, Lygodium japonicum,* and *Mohria Caffrorum.* Miss TWISS (14), investigating *Aneimia hirsuta, A. Phyllitidis,* and *Lygodium circinatum,* noted the points made by these workers. In addition, she found that the rhizoid in *Aneimia* and *Lygodium* is produced from the smaller cell formed in the first division of the germinating spore; that the apical cell is terminal and persistent in *Lygodium,* but is lateral and replaced early by a group of initial cells in *Aneimia;* and that the prothallium is heart-shaped, unsymmetrical at first in *A. Phyllitidis* and *L. circinatum,* and permanently so in *A. hirsuta.*

Methods

The spores of *Lygodium palmatum* (Bernh.) Sw. were collected in a somewhat swampy woods near South Hadley, Massachusetts, in November 1920, and March 1921. Those from the first collection were planted on peat, and on pieces of broken flower pots; those of the second collection were planted on the same substrata, and also on sphagnum, steam-sterilized soil, distilled water, and on Knop's solution. The formula for the Knop's solution used is as follows: to one liter of distilled water are added 9.25 gm. potassium nitrate, 0.25 gm. potassium phosphate, 0.25 gm. magnesium sulphate, 1.0 gm. calcium nitrate, and a trace of iron chloride. The calcium nitrate should be dissolved and added separately. The liquid of this strength was used as a nutrient solution for all pro-

thallia grown. The growth of fungi was kept down by the use of potassium permanganate of the strength suggested by Miss TWISS. The media and the containers were sterilized, and the sporangia were washed in potassium permanganate before the spores dropped. The prothallia were grown in covered dishes at room temperature (17°–19° C.), some at a north window with no direct sunlight, others at an east window where they were exposed to sunlight for from two to three hours a day. Cultures of *L. circinatum* (Burm.) Sw. (*L. dichotomum* Sw.) were also made. The fertile fronds were obtained from the greenhouses of Harvard University. The number of spores of *L. palmatum* which dropped from the sporangia gathered in the early collection was small, and although the spores germinated, the work, excepting that on the archegonia, was done with the later collection. The early stages of development were obtained from crock cultures, the first divisions being made visible by clearing in glycerine or potassium hydroxide, of which the former proved the more satisfactory. With this exception, all the work was done with living material. Spores were kept at three temperatures, 7° C., 17°–19° C., and 25°–27° C., with the result in each case that germination still took place at the end of eleven months. The spores planted at the end of three months produced less vigorous prothallia than those planted directly after collection. This was true of all later cultures. That each successive planting produced prothallia increasingly less vigorous was shown by their smaller size, and by their slower growth. The rate and percentage of germination were decreased. Practically all the spores planted directly after collection germinated, but of those which were kept seven months, fifty per cent began to crack only after twelve days.

Observations

The spores are cinnamon colored in mass. They are tetrahedral, and the wall is opaque. They are 57μ in diameter, measured from the base to the apex. This exceeds measurements of the spores of *Pteris tremula*, which are of the same type, by 14μ. The surface is covered with short, blunt protuberances which give the spore a rough appearance. The cultures placed in the north window grew best, the others lagging behind a few days in all stages up to

the formation of the sex organs. The prothallia planted on peat developed at about the same rate as those on soil; those planted on sphagnum more slowly; and the development of those on Knop's solution was slowest of all.

The outer layer of the spore wall cracks in four to six days (fig. 1), and the first wall, which is parallel to the plane of the crack and within the spore wall (fig. 2), divides the germinating spore into a basal cell which remains within the spore wall, and a terminal cell which protrudes slightly. The membrane of the terminal cell when treated with a weak solution of water-soluble aniline blue or of resorcin blue, stains in the same way as the inner membrane of the spore, and seems to be continuous with it. The second division of the spore (figs. 3, 4) occurs in a plane more or less nearly perpendicular to the first, dividing the terminal cell into a rhizoidal cell (*rh*) and a primary prothallial cell (*pp*). The latter contains a great abundance of chloroplasts, contrasting with the rhizoidal cell which has no chloroplasts and is transparent. The primary prothallial cell usually grows more rapidly than the rhizoidal cell. The third wall (figs. 5, 6) divides the prothallial cell into a larger cell, the prothallium initial (*pi*), and a smaller cell (*w*) which appears nearly wedge-shaped (fig. 7), and which ordinarily does not divide further. The young prothallium, therefore, is formed from the prothallium initial (figs. 9, 10). I have found a few cases of branched prothallia. In one of these cases, the wedge-shaped sister cell of the prothallium initial had developed into a short, protruding row of cells (fig. 12 *f*), and the prothallium initial, by a longitudinal division, had formed two papillae, one of which (p^1), noticeably larger than the other (p^2), had developed into a young prothallium. This arrangement, in appearance though not in origin, resembles that found in *L. circinatum* (**14**). Fig. 11 shows an individual of the type just described, in which, however, the wedge-shaped cell had not divided. Miss WUIST (**15**), working on branched prothallia of the Polypodiaceae, says that there seems to be a definite relation between branching and nutrition. Such branching as I found in my cultures might possibly be due to nutritive conditions. The prothallia described by Miss WUIST, however, form branches by division of any cell, including the end cell, of a

filament; or from the margin or apex of an older prothallium. On the other hand, the branching I have found is confined to the divisions of the primary prothallial cell. I did not follow the further development of these prothallia, and so do not know what may become of the branches. It is possible that they produce the irregular margins (fig. 20) found in some older prothallia.

The young prothallium is formed from the prothallium initial in one of three ways, just as Miss TWISS has described for *L. circinatum*. The most general method is by horizontal (fig. 8) and longitudinal walls producing two rows of cells placed side by side (fig. 16). A second type is marked by the formation of oblique walls from the very first (figs. 7, 9). In the third type the first few divisions are transverse, resulting in the formation of a filament of a single row of cells (fig. 14). The prothallia produced by the first two types of development are more than one cell thick from the very beginning (fig. 15), and often are three or four cells in thickness (fig. 13), forming a cell mass in the very early stages of growth. This cell mass, which is so characteristic of the very young prothallia of *L. palmatum*, is not described for *L. circinatum* by Miss TWISS. The results obtained in my cultures of *L. circinatum* confirm her descriptions. KNY (**11**) and CAMPBELL (**6, 8**), working on *Osmunda*, and LUERSSEN (**12**) on *Todea*, report that a cell mass (or body) is formed when the spores germinate. CAMPBELL states further that this may not always be the case, for in *O. Claytoniana* a filamentous protonema may be formed, which broadens into a plate of cells, as in the Polypodiaceae described by PRANTL (**13**). In the development of the cell mass of *Osmunda*, the first wall divides the spore into a smaller cell, which, protruding from the spore, elongates to form the first rhizoid, and a larger prothallium mother cell which by further divisions forms the prothallium. The germination is bipolar, that is, the growth of the prothallium is in a direction opposite to that of the rhizoid. CAMPBELL and LUERSSEN figure the very young prothallium as a compact tissue often at least two cells thick. There is a resemblance here to the cell mass in *L. palmatum*, but in the latter the germination has never been found to be bipolar. This formation of a cell mass at an early stage, which is found in *L. palmatum*, has not been

reported for the Schizaeaceae, and it may be considered a new type for the prothallia of homosporous leptosporangiate ferns.

Filaments consisting of a single row of cells (figs. 17, 18) with little chlorophyll were found in crowded cultures, and in some cultures grown on sphagnum. One filament was formed from each spore. BAUKE (3) reported such a development of filaments for *L. japonicum*. The first one or two basal cells in such a filament are unusually long, and occasionally the terminal cell is found to have divided longitudinally (fig. 18), thus initiating the development of a plate of cells. I did not follow the later history of these individuals. They do not represent the ordinary development, which is characterized by the formation from the prothallium initial of a distinct cell mass.

In about two weeks after germination has begun, an apical cell may appear, which is terminal (figs. 15, 16), as BAUKE (3, 4) and Miss TWISS have reported for *Lygodium*. It is remarkably persistent, often being present as late as the time of the formation of the first antheridia (fig. 20), but it is finally replaced by a group of initials. Within about four weeks the cell mass becomes broadened into a thallus one cell in thickness (fig. 19). This very soon becomes heart-shaped (figs. 20, 21), with a midrib two to seven cells thick, running backward from the apical notch. The wings, or lobes, of the prothallium in *L. palmatum* are substantially symmetrical. The unequal lobing reported for young prothallia of *L. japonicum* by BAUKE (3), and for those of *L. circinatum* by Miss TWISS, was not conspicuous in the prothallia of my cultures of *L. circinatum*. Miss TWISS also found the prothallia of *A. hirsuta* and *A. Phyllitidis* to be asymmetrical. In *L. palmatum* a slight variation in the size of the lobes of the prothallia (figs. 19, 20) was not infrequently found, but such differences in size did not seem sufficiently marked to be significant.

In general there are no hairs on the prothallia. HEIM (9) reports that there are no hairs of any kind on the prothallia of *Lygodium*, but that in *Aneimia* and *Mohria*, on all parts of the thallus, there are characteristic "nierenförmige Haare," containing tannic acid, and inclined toward the apex. He gives no figures. BAUKE (2) found short papillae, at relatively wide distances,

exclusively on the margin of the prothallia of *A. Phyllitidis*, and these were almost always bent toward the apical region of the thallus. His figures show them to be in general one-celled structures, separated in each case by a wall from the surface cell. In *Mohria Caffrorum*, however, these hairs are egg-shaped, unicellular, and are ordinarily found on the surface of the thallus. BURCK and Miss TWISS do not mention or figure hairs. I have found two-celled hairs on two specimens of *L. palmatum*. On one of these, a young prothallium bearing both archegonia and antheridia (fig. 24), there were two hairs at the notch, opposite each other and bending backward toward the row of initial cells (fig. 23). Each is composed of a basal cell and a longer, slightly swollen, terminal cell. These two hairs resemble those which I have seen, and which are described by CAMPBELL (7) as short and glandular, accompanying the archegonia in *Gleichenia*. In another case (fig. 22) a single hair appeared near the apical notch on a regenerated thallus (fig. 36) bearing archegonia. I did not find hairs on the prothallia of *L. circinatum*, and the two cases just described for *L. palmatum* should hardly be considered significant. It is quite possible that, as a regular thing, hair formation is restricted to the two genera *Aneimia* and *Mohria*, as HEIM states.

The antheridia in my cultures appear when the prothallia are about two or three months old, and are found on the cushion just behind the apical notch (figs. 20, 25). Archegonia are found in the same region on other prothallia which are three to four months old (fig. 26). Occasionally prothallia are found bearing sex organs of both kinds, the archegonia at the notch, the antheridia behind them (fig. 24). HEIM reported that in his typical cultures of *L. japonicum* archegonia appeared first, and that the young antheridia, situated on both sides of the midrib in the region surrounding the apical notch, appeared later, and were still present after fertilization. Miss TWISS, however, finds that the antheridia develop first in *L. circinatum*, followed by the appearance of archegonia, which may be on the same or on separate prothallia. This corresponds to the condition found in *L. palmatum*. My cultures show that a prothallium may produce an enormous number of archegonia. On prothallia six to eight months old

(fig. 33), from two to three hundred archegonia may be found all over the wide cushion, the youngest being nearest the apical notch. Prothallia in crowded cultures show no tendency to develop ameristic male prothallia, as is the case in the Cyatheaceae and the Polypodiaceae (BAUKE 1, PRANTL 13). The male prothallia of these two families generally lack an apical growing region and a midrib. They are irregular branching threads, or plates of cells, which bear antheridia exclusively. No prothallia of this sort were found in my cultures, but many individuals of the type shown in fig. 27 were found in cultures which were crowded, or which had grown under conditions of restricted nutriment. The prothallium shown in fig. 27 had reached the apical cell stage when the apical cell grew out into a filament bearing antheridia. Another type of male prothallium was found in these same cultures, and also included nearly every individual of those grown on sphagnum, although the latter were not particularly crowded, and certainly not starved. In these cases (fig. 28) a prothallium bearing many antheridia had formed an apical region when the group of initial cells grew out into a ribbon-like process, bearing antheridia on the margins and on both surfaces. In some cases I observed an apical cell on such a ribbon-like process; in others the apical cell had been replaced by a group of initials. I have found prothallia also of this latter type bearing archegonia and rhizoids (fig. 36). Such prothallia were abundant in cultures which had been moved from place to place, and had been carried on peat from Massachusetts to Wisconsin. The particular prothallium figured was the only one of this type found bearing a hair. These modifications in development are undoubtedly due to adverse conditions, of which a change in nutrition may be one. Since an apical growing region and rhizoids are present, these prothallia may be regarded as regenerated types.

Regeneration occurs not infrequently. I have observed several instances in which the cells at the tip of a young prothallium were injured. In each case the injury had taken place just before, or just at the time of, the formation of the apical cell, and the filament had grown from the thallus, had developed an apical region, and had broadened into a prothallium with rhizoids. If the

marginal cells at the tip of the thallus were destroyed, the filament came from some lateral cell (figs. 29, 30); if the cells just behind the tip were affected, the apical cell itself grew out (fig. 31).

The prothallia are positively phototropic. Since they were grown by a window, they were subjected to unilateral illumination. The prothallia, especially the younger heart-shaped ones, leaned toward the source of illumination, standing at an angle from the medium of at least 45°, the wings extended stiffly. As the prothallia grew older there was a marked ruffling of the wings (figs. 26, 33, 34), with the result that they curved over the dorsal surface of the thallus until they touched each other (fig. 32). The portions of a prothallium which were shaded and kept moist in this way, or which were partially covered by another prothallium, produced rhizoids and archegonia on the dorsal surface, although these organs are most commonly borne on the ventral side of the midrib. BAUKE (**3**) reports that rhizoids and archegonia are produced on both sides of the thallus when it is standing perpendicularly.

Branching of the midrib has been reported by BAUKE (**3, 4**) as occurring often in *L. japonicum* and also in *Balantium antarcticum*. I found that on every healthy prothallium of *L. palmatum* which had grown to the age of six months there were at least two apical regions (fig. 33), each at the end of a branch of the midrib. One prothallium, eight months old, has produced four apical regions (fig. 34), with corresponding branchings of the cushion.

Summary

1. *Lygodium palmatum* shows a new type of germination. The first division of the spore is by a wall horizontal to the crack. This is followed by a longitudinal division of the outer cell, forming a rhizoidal cell and a primary prothallial cell. The prothallial cell is the first to protrude from the spore.

2. A cell mass is formed immediately in the young prothallium, preceding the formation of a plate of cells. A terminal apical cell appears early, often persisting until the development of antheridia. Eventually the apical cell is replaced by a group of initial cells.

3. The wings, which are one cell thick, are substantially symmetrical from the start.

4. The midrib of the heart-shaped thallus is thick and broad, branching when the prothallium is about six months old.

5. The sex organs and rhizoids are usually produced on the ventral surface, but they may occur on the dorsal surface when this is kept shaded and moist.

6. Under adverse conditions, male prothallia with an apical region are produced by an outgrowth of an apical cell or of the initial cells.

7. Regeneration occurs when the terminal cells of the thallus are injured.

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LITERATURE CITED

1. BAUKE, H., Entwickelungsgeschichte des Prothalliums bei den Cyatheaceae, verglichen mit derselben bei den anderen Farrenkräutern. Jahrb. Wiss. Bot. **10**:49–116. *pls. 6–10.* 1876.
2. ———, Beiträge zur Keimungsgeschichte der Schizaeaceen. Jahrb. Wiss. Bot. **11**:603–650. *pls. 38–41.* 1878.
3. ———, Zur Kenntniss der sexuellen Generation bei ben Gattungen *Platycerium*, *Lygodium*, und *Gymnogramme*. Bot. Zeit. **36**:753–760; 769–780. 1878.
4. ———, Aus dem botanischen Nachlasse von Bauke. Beilage zur Bot. Zeit. **38**: *pls. 1–6* (following p. 904). 1880.
5. BURCK, W., Vorläufige Mitteilung über die Entwickelungsgeschichte des Prothalliums von *Aneimia*. Bot. Zeit. **33**:499–501. 1875.
6. CAMPBELL, D. H., On the prothallium and embryo of *Osmunda Claytoniana*, L., and *O. cinnamomea*, L. Ann. Botany **6**:50–94. *pls. 3–6.* 1892.
7. ———, The prothallium of *Kaulfussia* and *Gleichenia*. Ann. Jard. Bot. Buitenzorg II. **7**:69–102. *pls. 7–14.* 1908.
8. ———, Mosses and ferns, 3d ed. New York. 1918.
9. HEIM, C., Untersuchungen über Farnprothallien. Flora **82**:329–373. *figs. 16.* 1896.
10. KNY, L., Entwickelungsgeschichte des Vorkeims der Polypodiaceen und Schizaeaceen. Report in Bot. Zeit. **27**:46. 1896.

11. KNY, L., Beiträge zur Entwickelungsgeschichte der Farrnkräuter. Jahrb. Wiss. Bot. 8:1–15. *pls. 1–3.* 1872.

12. LUERSSEN, C., Zur Keimungsgeschichte der Osmundaceen, vorzüglich der Gattung *Todea.* Mittheilungen aus dem Gesammtgebiete der Botanik (Schenk und Luerssen) 1:460–477. *pls. 13, 14.* 1874.

13. PRANTL, K., Über die Anordnung der Zellen in flächenförmigen Prothallien der Farne. Flora 61:497–505; 529–539; 545–556. *pls. 2, 3.* 1878.

14. TWISS, EDITH M., The prothallia of *Aneimia* and *Lygodium.* BOT. GAZ. 49:168–181. *pls. 10, 11.* 1910.

15. WUIST, E. D., Branched prothallia in the Polypodiaceae. Bull. Torr. Bot. Club 43:365–383. *figs. 15.* 1916.

EXPLANATION OF PLATES IV–VI

All figures except fig. 32 were drawn at table level with the aid of an Abbé camera lucida, using Leitz oculars and objectives in combinations to give the magnifications listed.

Reference letters: *a*, apical region; *ap*, apical cell; *bc*, basal cell; *f*, filament growing from wedge-shaped cell; *h*, hair; *pi*, prothallium initial; p^1, p^2, papillae; *pp*, primary prothallial cell; *pro*, first prothallium; *rh*, rhizoid; *w*, wedge-shaped cell.

PLATE IV

FIG. 1.—Lateral view of germinating spore, showing protruding terminal cell; ×240.

FIG. 2.—First division of spore; ×240.

FIG. 3.—Division of terminal cell, as seen in apical view; ×240.

FIG. 4.—Same, seen in lateral view; ×210.

FIG. 5.—Lateral view of division of primary prothallial cell; ×210.

FIG. 6.—Same, in apical view; ×240.

FIGS. 7, 8.—Divisions of prothallium initial; ×210.

FIG. 9.—Young prothallium; ×210.

FIG. 10.—Young prothallium with outer layer of spore wall removed, showing development of prothallium from prothallium initial; ×170.

FIG. 11.—Branched prothallium; ×170.

FIG. 12.—Branched prothallium; ×210.

FIG. 13.—Lateral view of prothallium, showing cell mass; ×57.

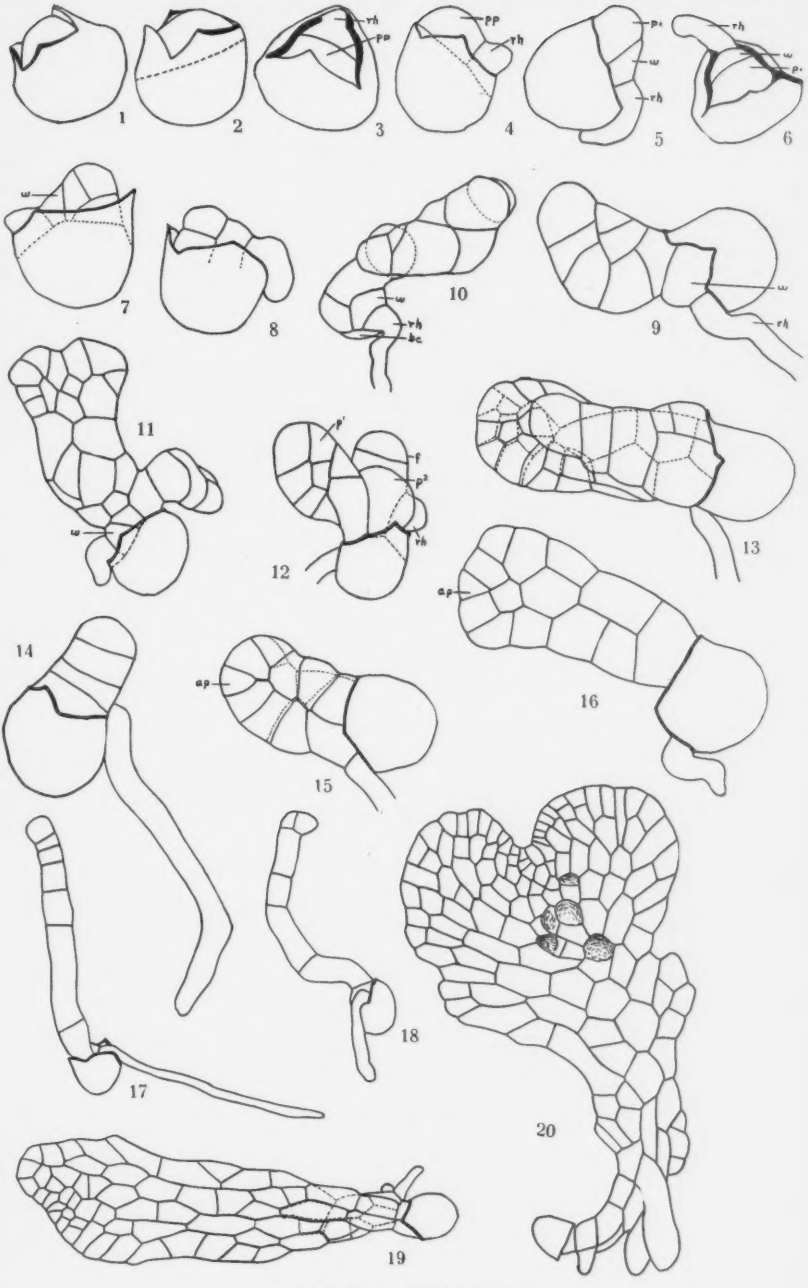
FIG. 14.—Stage corresponding to that of figs. 7 and 8, prothallium initial having developed another way; ×170.

FIGS. 15, 16.—Prothallia, each with apical cell; ×210.

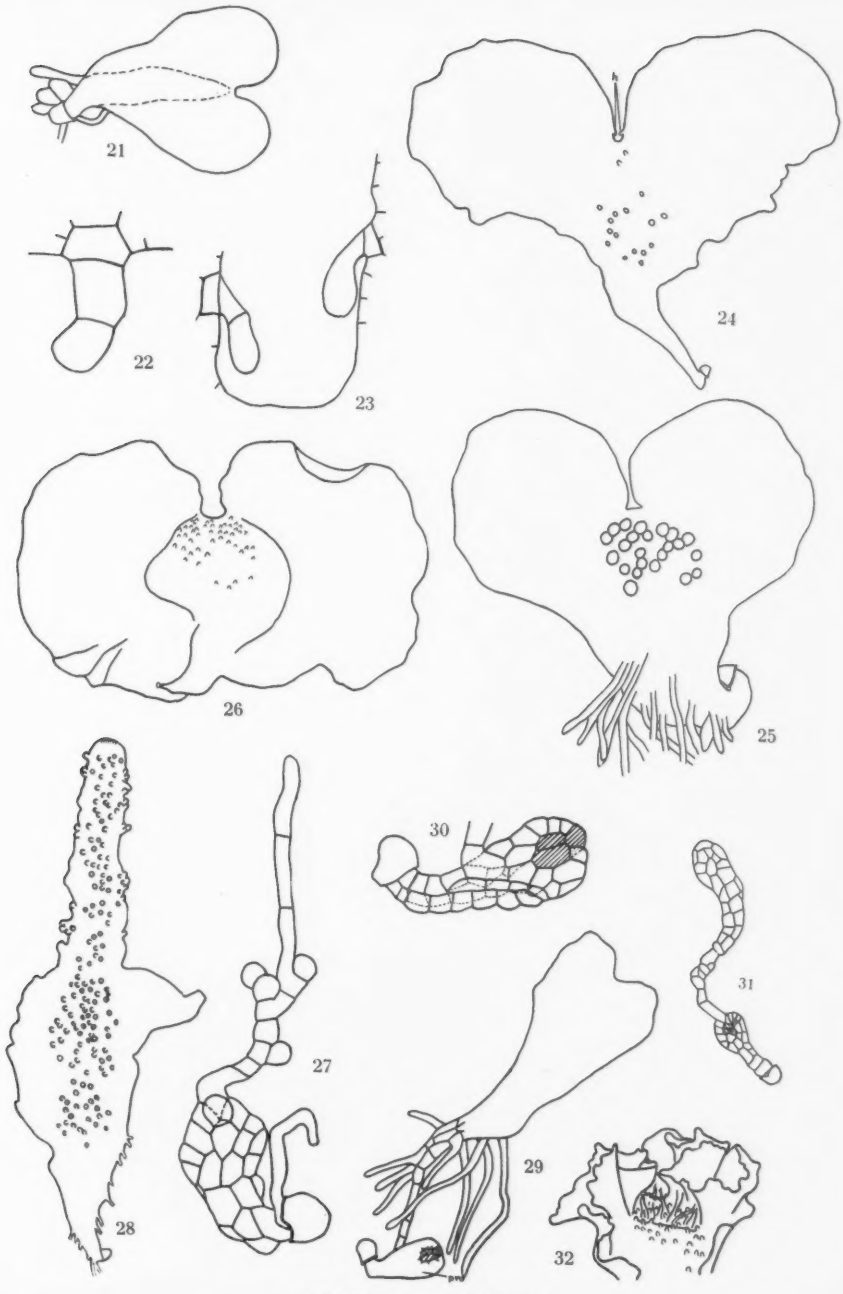
FIGS. 17, 18.—Filamentous prothallia; ×105.

FIG. 19.—Plate of cells replacing cell mass; ×87.

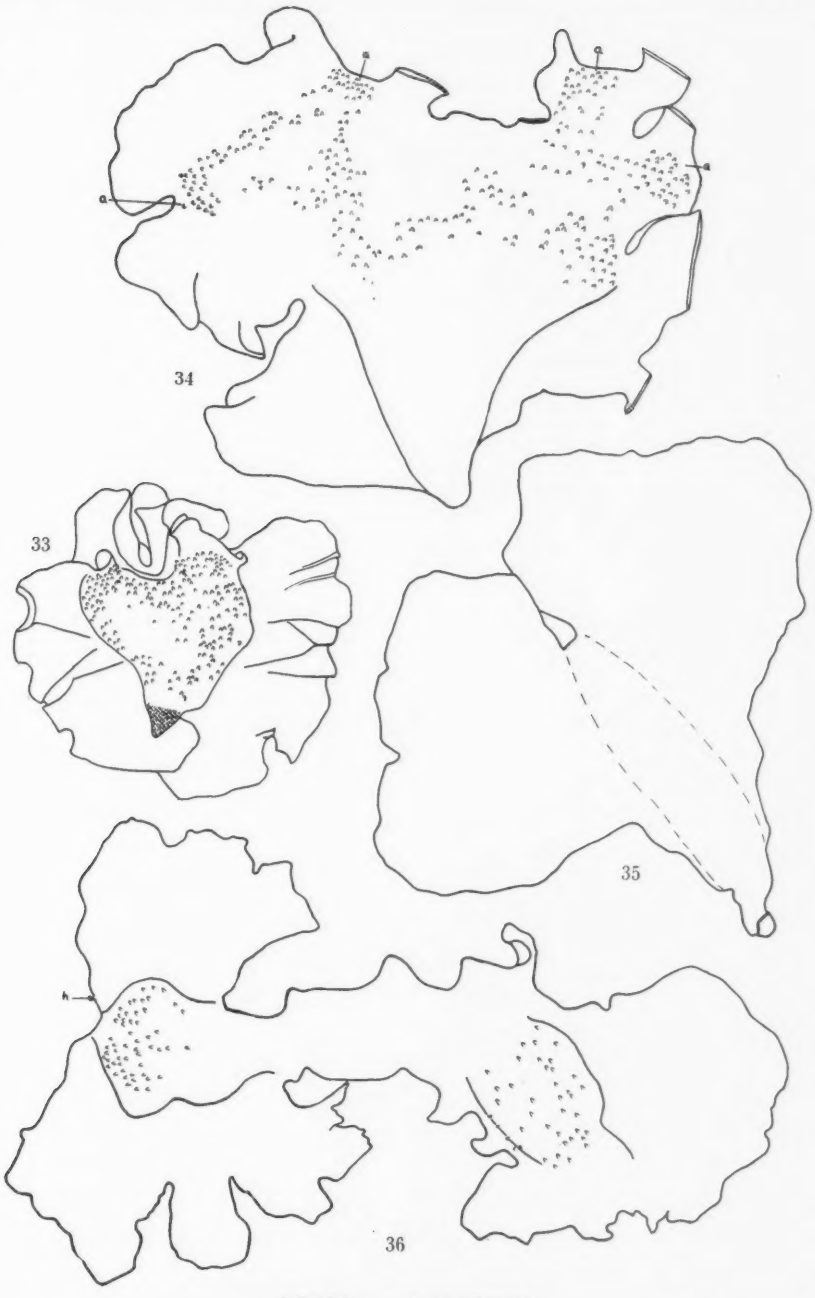
FIG. 20.—Ventral view of prothallium bearing antheridia; ×87.



ROGERS on LYGODIUM



ROGERS on LYGODIUM



ROGERS on LYGODIUM

PLATE V

FIG. 21.—Heart-shaped prothallium with midrib; ×41.5.

FIG. 22.—Hair, magnified from fig. 36*h*, ×210.

FIG. 23.—Hairs magnified from fig. 24*h*; ×210.

FIG. 24.—Prothallium bearing archegonia, antheridia, and two hairs; ×16.

FIG. 25.—Prothallium bearing antheridia; ×19.

FIG. 26.—Prothallium bearing archegonia; ×7.5.

FIG. 27.—Male prothallium of type found in crowded cultures; ×87.

FIG. 28.—Male prothallium with regenerative outgrowth from apical region; ×57.

FIG. 29.—Regenerated prothallium; ×14.

FIG. 30.—Enlarged from fig. 29 *pro*, showing region of regeneration; ×87.

FIG. 31.—Regenerated prothallium; ×41.5.

FIG. 32.—Dorsal portion of prothallium, looking along surface toward apical notch beneath ruffled wings, showing archegonia and rhizoids.

PLATE VI

FIG. 33.—Old prothallium with branched midrib bearing many archegonia; ×7.5.

FIG. 34.—Old prothallium with four apical regions; ×7.5.

FIG. 35.—Typical heart-shaped prothallium; ×19.

FIG. 36.—Regenerated prothallium bearing archegonia and a hair; ×7.5.

DEHYDRATION OF CERTAIN PLANT TISSUES[1]

VICTOR R. BOSWELL

(WITH SEVEN FIGURES)

The literature of horticulture contains much evidence to show that injury from cold is associated with withdrawal of moisture from the cells of the plant, and that hardiness is correlated with ability of the plant to retain water. If hardy tissue will not give up its water as readily as tender tissue when heated, it should show a similar power of resistance to any force tending to withdraw water from the cells upon freezing. This investigation was made to determine (1) the relative water-retaining capacity of the tissues of plants subjected to different treatments and of known degrees of hardiness, and (2) whether there is close correlation between the relative amounts of free and unfree water in such tissues and their rate of drying.

Materials and procedure

The investigation was made with the use of cabbage and tomato leaves of plants of known degrees of hardiness. Care was taken to secure only leaves of uniformly good condition, and of as nearly the same age as possible. A few leaves were taken from the plants for each sample, immediately placed in large shallow weighing bottles with tight glass covers, and weighed. As soon as possible after weighing the leaves were carefully spread in an oven on a shelf which was covered with wire gauze. A temperature of approximately 60° C. was maintained. The samples were dried fifteen minutes and then removed, quickly placed in the bottles, covered, cooled, and again weighed. The leaves were then spread on the gauze as before, and the process was repeated until a weight very near constant was reached. In some of the experiments duplicate samples were taken from each lot of plants. These samples were

[1] Contribution from the Department of Horticulture of the Agricultural Experiment Station of the University of Missouri.

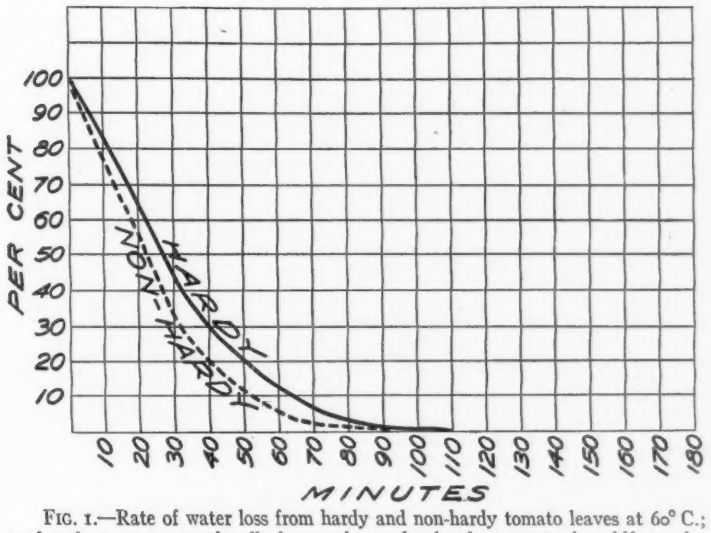


FIG. 1.—Rate of water loss from hardy and non-hardy tomato leaves at 60° C.; tender plants grown continually in greenhouse, hardy plants grown in coldframe for two weeks.

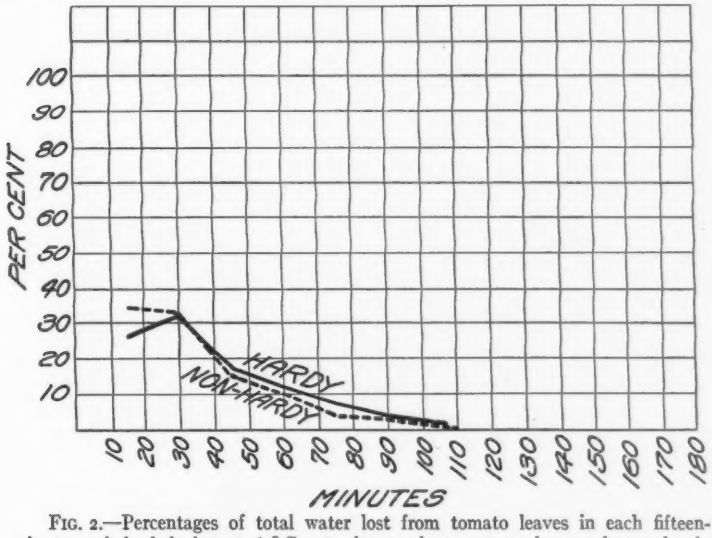


FIG. 2.—Percentages of total water lost from tomato leaves in each fifteen-minute period of drying at 60° C.; tender specimens grown in greenhouse, hardy specimens grown in coldframe for two weeks.

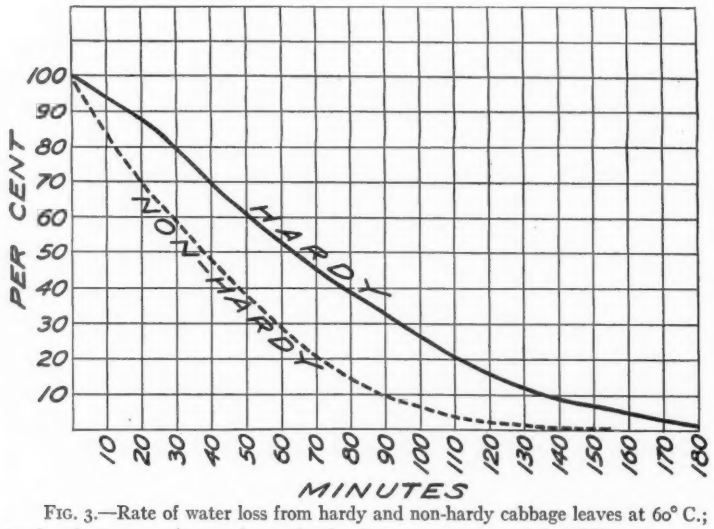


FIG. 3.—Rate of water loss from hardy and non-hardy cabbage leaves at 60° C.; tender plants grown in greenhouse, hardy plants grown one week in coldframe.

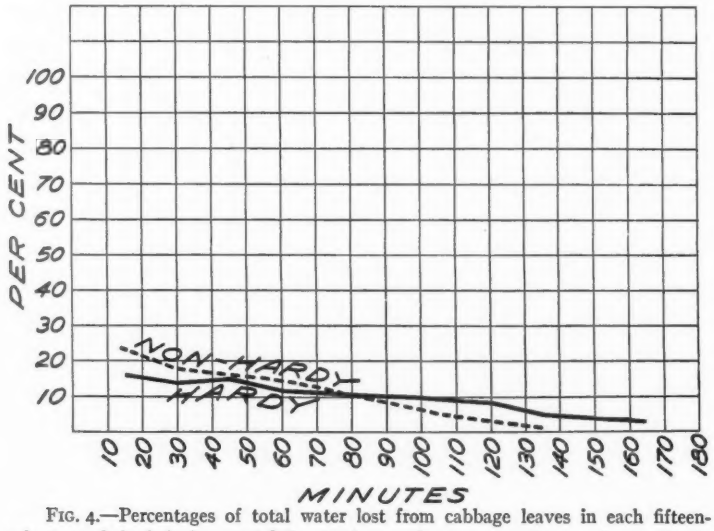


FIG. 4.—Percentages of total water lost from cabbage leaves in each fifteen-minute period of drying at 60° C.; tender specimens grown in greenhouse, hardy specimens grown in coldframe for one week.

heated alternately, that is, while one was drying in the oven the other was being cooled and weighed.

Data

The results of the dehydration experiments with cabbage and tomato leaves are presented in tables I to V, and illustrated graphically in figs. 1 to 7. Each table gives the percentage of the total

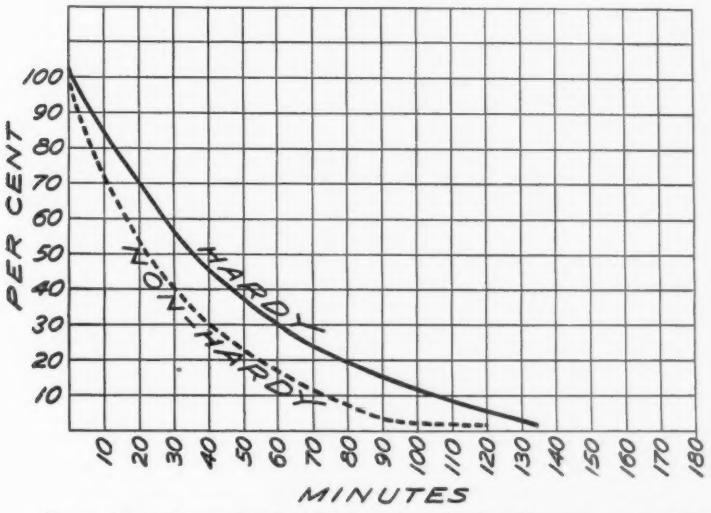


FIG. 5.—Rate of water loss from hardy and non-hardy cabbage leaves at 70° C.; tender plants grown in greenhouse in very moist soil, hardy plants grown in dry soil.

water content of the tissue removed in the particular fifteen minute interval indicated, and the percentage of the total water which had been removed in the entire drying period, the results being additive. Graphs in figs. 1, 3, 5, 6, and 7 were plotted from the data in tables I and III, and V, and arranged so as to show percentages of water retained after drying. Those in figs. 2 and 4 are smoothed curves representing the averages of determinations from duplicate samples heated alternately for thirty minutes each, with the exception of the first period, which was but fifteen minutes. Curves showing the amounts of water retained by the different lots

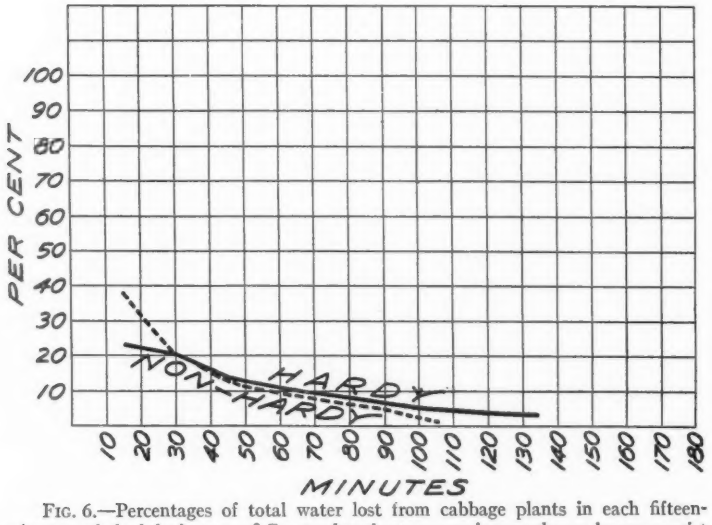


FIG. 6.—Percentages of total water lost from cabbage plants in each fifteen-minute period of drying at 70° C.; tender plants grown in greenhouse in very moist soil, hardy plants grown in dry soil.

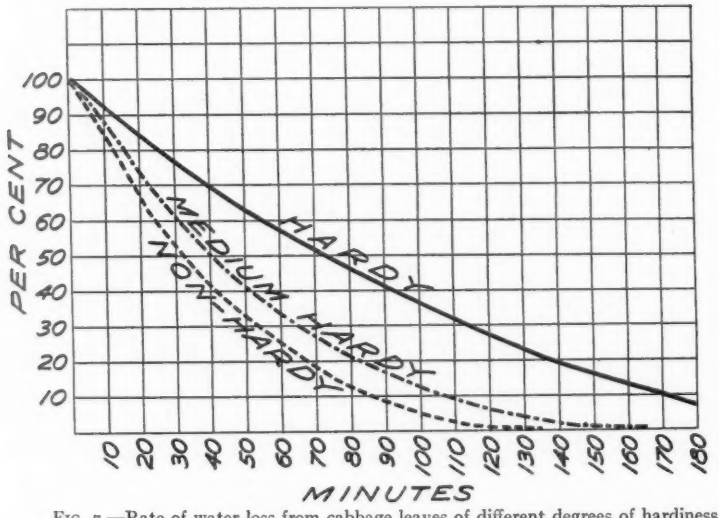


FIG. 7.—Rate of water loss from cabbage leaves of different degrees of hardiness at 60° C.; tender plants watered with tap water, medium hardy plants with N/10 $NaNO_3$ solution, and hardy plants with N/10 NaCl solution; all grown in greenhouse.

when subject to continued drying would parallel those showing the rate of water loss.

TABLE I

DEHYDRATION OF TOMATO LEAVES*

MINUTES	PERCENTAGE OF TOTAL WATER REMOVED		PERCENTAGE OF TOTAL WATER REMOVED PER PERIOD	
	Greenhouse (tender)	Coldframe (hardy)	Greenhouse (tender)	Coldframe (hardy)
15	34.77	26.46	34.77	26.46
30	68.11	57.91	33.34	31.45
45	83.99	75.34	15.88	17.43
60	94.27	87.85	10.28	12.51
75	97.94	95.57	3.65	7.72
90	99.34	99.02	2.40	3.45
105	99.59	99.52	0.25	0.50
120	99.62	99.61	0.03	0.09
135	99.64	99.63	0.02	0.02

* Tender plants were grown in the greenhouse in wet soil; hardy plants of the same age as the tender were grown two weeks in the coldframe in early March.

TABLE II

DEHYDRATION OF CABBAGE LEAVES*

MINUTES	PERCENTAGE OF TOTAL WATER REMOVED				PERCENTAGE OF TOTAL WATER REMOVED PER PERIOD			
	Greenhouse (tender)		Coldframe (hardy)		Greenhouse (tender)		Coldframe (hardy)	
	A	B	A	B	A	B	A	B
15	21.53	.....	8.92	.....	21.53	.....	8.92	.....
30	.....	42.71	.....	17.43	.....	42.71	.....	17.43
45	54.20	.....	36.83	.....	32.67	.....	29.54	.....
60	.....	75.32	.....	47.56	.....	32.66	.....	30.13
75	79.08	.....	55.23	.....	24.88	.....	18.40	.....
90	.....	93.32	.....	68.23	.....	18.00	.....	20.67
105	93.12	.....	74.58	.....	14.04	.....	19.35	.....
120	.....	99.66	.....	86.01	.....	6.34	.....	17.78
135	98.11	.....	87.47	.....	4.99	.....	12.89	.....
150	.....	99.83	.....	94.88	.....	0.17	.....	8.81
165	99.61	.....	94.73	.....	0.50	.....	7.26	.....
180	.....	99.83	.....	98.88	.....	0.00	.....	4.00
195	99.74	.....	97.90	.....	0.13	.....	3.15	.....
210	.....	99.83	.....	98.88	.....	0.00	.....	0.00
225	.....	.....	99.60	.....	.....	.....	1.70	.....
240	.....	.....	.....	99.74	.....	.....	.....	0.66
255	.....	.....	99.70	.....	.....	.....	0.10	.....

* Tender plants were grown in the greenhouse in wet soil; hardy plants were grown in the coldframe for one week in March.

TABLE III

DEHYDRATION OF CABBAGE LEAVES*

MINUTES	PERCENTAGE OF TOTAL WATER REMOVED		PERCENTAGE OF TOTAL WATER REMOVED PER PERIOD	
	Greenhouse (tender)	Coldframe (hardy)	Greenhouse (tender)	Coldframe (hardy)
15	43.72	26.51	43.72	26.51
30	73.98	53.31	20.26	26.80
45	84.64	66.68	10.66	13.37
60	91.85	77.22	7.21	10.54
75	96.40	88.62	4.55	11.40
90	98.46	93.97	2.06	5.35
105	99.52	97.78	1.06	3.81
120	99.77	98.62	0.25	0.84
135	99.83	99.46	0.06	0.84
150	99.85	99.79	0.02	0.23
165	99.87	99.81	0.02	0.02

* Tender plants were grown in the greenhouse in wet soil; hardy plants of the same age as the tender were grown ten days in the coldframe in March.

TABLE IV

DEHYDRATION OF CABBAGE LEAVES*

MINUTES	PERCENTAGE OF TOTAL WATER REMOVED				PERCENTAGE OF TOTAL WATER REMOVED PER PERIOD			
	Wet grown (tender)		Dry grown (hardy)		Wet grown (tender)		Dry grown (hardy)	
	A	B	A	B	A	B	A	B
15	39.20	.....	22.90	.....	39.20	.....	22.90	.....
30	.....	61.40	.....	45.70	.....	61.40	.....	45.70
45	73.50	.....	57.90	.....	34.30	.....	35.00	.....
60	.....	84.30	.....	70.00	.....	22.90	.....	29.30
85	98.40	.....	86.40	.....	24.90	.....	28.50	.....
100	.....	95.00	.....	86.40	.....	10.70	.....	16.40
115	100.00	.....	96.00	.....	1.60	.....	9.60	.....
130	.....	99.90	.....	97.00	.....	4.90	.....	11.00

* Tender plants were grown in greenhouse in wet soil; hardy plants were grown in greenhouse in dry soil.

Discussion

The tables show that in every case the hardy tissue lost water less rapidly than the tender tissue. The differences in the rate of loss varied greatly, depending upon the kind of plant and the treatment to which it had been subjected. Fig. 1, representing the results obtained with the tomato, shows that the plants sub-

jected to hardening treatment lose their water nearly as rapidly as those unhardened; and the two curves run close together. Furthermore, the loss of water is rapid, over 90 per cent of the total being removed in one hour, and over 99 per cent in ninety minutes. The hardening treatment had not resulted in the development of any considerable water-retaining power in the tomato leaves. On the other hand, fig. 3, representing similar determinations for cabbage, shows a decided difference in rate of water loss from hardened and

TABLE V

DEHYDRATION OF CABBAGE LEAVES*

MINUTES	PERCENTAGE OF TOTAL WATER REMOVED			PERCENTAGE OF TOTAL WATER REMOVED PER PERIOD		
	H_2O	$NaNO_3$	NaCl	H_2O	$NaNO_3$	NaCl
15	23.82	19.70	12.13	23.82	19.70	12.13
30	46.71	38.68	24.29	22.89	18.98	12.16
45	62.74	54.80	34.68	16.03	16.12	10.39
60	74.81	66.27	43.13	12.07	11.47	8.45
75	84.67	76.42	51.99	9.86	10.15	8.86
90	91.59	83.20	59.36	6.91	6.78	7.37
105	96.32	89.27	66.79	4.73	6.07	7.43
120	98.73	93.94	73.34	2.41	4.67	6.55
135	99.68	97.57	78.87	0.95	3.63	5.53
150	99.93	99.37	84.48	0.25	1.80	5.61
165	99.95	99.87	88.91	0.02	0.50	4.43
180	99.96	99.95	94.68	0.01	0.07	5.77
210	100.00†	100.00†	99.65†	0.04†	0.05†	4.97†

* All plants were grown in the greenhouse, but were watered with tap water, or with N/10 sodium nitrate and sodium chloride as indicated at the head of the columns.

† Thirty minute period.

unhardened tissue, even though the hardening treatment to which these plants were subjected was less severe than that afforded the tomato plants. If figs. 1 and 3, or figs. 2 and 4 are compared, it will be noted that the rate of water loss in the tender cabbage is less than that in the hardened tomato plants. These relative rates of water loss parallel more or less closely their known degrees of cold resistance.

Still more striking differences are shown in table V and fig. 7. The plants from which these curves were drawn were grown under uniform conditions in the greenhouse. One was watered with tap water, one with N/10 sodium chloride, and one with N/10 sodium

nitrate. These treatments resulted in plants of very different degrees of hardiness, as has been shown elsewhere.[1] The drying curve for the tender plants closely parallels that for the tender plants shown in fig. 3. The second lot, which were hardier, lost water at a slower rate. The third lot, considerably hardier, lost water at a still slower rate. The figures show conclusively that there is a close correlation between the degree of hardiness and the rate of water loss.

Other interesting points are brought out by fig. 6, showing the percentage of total water contained in the tissue which was removed in each period of drying. In every case the curve for the hardy tissue starts lower than that for tender tissue, crosses the curve for tender tissue, and at the end of the drying has the higher value. This shows that the hardy tissue loses its water more slowly in the early periods of drying, and consequently has more to lose in the later periods. The curve for the hardy tissue falls less rapidly, and in very hardy tissues in almost a horizontal line, indicating that its water is held more tenaciously than in the tender tissue. The water that is held so firmly is presumably in colloidal combination, while that which passes off early in the drying process is probably free water. In the case of the cabbage tissues used in this investigation, thirty to forty minutes' drying at 60° C. removed practically all of the free water, as determined by dilatometer experiments; and the differences in the heights of the curves at that point indicate approximately the relative amounts of colloidal water. Fig. 6 is not directly comparable with the others, for the drying temperature was higher than for the other determinations.

The results suggest that the method of drying described in this paper may be used to estimate the relative amounts of free and unfree water in plant tissues, and that the comparative rates of water loss under uniform drying conditions are a measure of the relative hardiness of certain kinds of plant tissues.

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[1] ROSA, J. T. Jr., Investigations on the hardening process in vegetable plants. Mo. Agric. Exp. Sta. Research Bull. 48, 97. 1921.

POT CULTURES WITH BARLEY IN SOIL FROM A LONG-TIME FERTILIZER EXPERIMENT[1]

A. R. C. HAAS

(WITH FOUR FIGURES)

The field trials conducted by the Citrus Experiment Station, Riverside, California, to test the effect of different fertilizers upon orange and lemon trees, have been running since 1907. The main features concerning the treatments and their effects on the trees have been presented by VAILE.[2] The question has been raised whether a cereal crop grown on these plots would show similar effects of the fertilizer treatment. It might be thought that a cereal plant, with its shorter period of growth, would respond in a manner different from that of a perennial tree. The present paper presents the results of barley cultures grown in soil taken from the different plots of this experiment.

Soil was taken with a spade from a dozen or more places on each plot, the composite sample amounting to several hundred pounds. After thorough mixing, three 15-inch pots, each containing from 100 to 125 pounds, were filled with the soil from each plot, thus giving a series of triplicate tests. This obviated the necessity of growing the plants in the field, with the consequent effects on the trees in the field trials. The last application of fertilizer previous to the time of drawing the samples was made in February 1920. The soil used in the present studies was taken from the plots on October 29, 1920. Beldi barley was planted November 6, 1920, and the crop was harvested May 7, 1921. When the seedlings were one to two inches high their number was reduced to 20 per pot. Throughout the growing period the plants were irrigated with distilled water.

[1] Paper no. 98, University of California, Graduate School of Tropical Agriculture and Citrus Experiment Station, Riverside, California.

[2] VAILE, R. S., Fertilizer experiment with citrus trees. Cal. Agric. Exp. Sta. Bull. 345. 1922.

In each case it was found, upon emptying the containers, that the roots were uniformly distributed throughout the mass of soil. The nitrate content of the soil at the time of planting the barley (table I) did not bear any strict relationship to the amounts of nitrogenous matter annually added to the plots. Nevertheless, the growth of the barley plants reduced the nitrate content of all samples to uniformly minimum amounts. This finding agrees

TABLE I

NITRATE CONTENT (P.P.M.) OF SOILS BEFORE AND AFTER GROWTH OF BARLEY PLANTS

Plot	At time of planting	At time of harvesting	Plot	At time of planting	At time of harvesting
A	16	3	L	14	2
B	7	2	M	8	2
C	32	2	N	21	4
D	12	3	O	50	4
E	21	3	P	13	3
F	42	4	Q	37	3
G	27	3	R	8	2
H	36	3	S	57	4
I	15	2	T	14	2
J	11	3	Field 2, Box Springs	21	4
K	10	3			

well with the results of STEWART and MARTIN,[1] who found that barley plants may greatly deplete the nitrate content of a soil. This may be of significance in the choice of barley as a winter cover-crop in orchards. A heavy growth of such a cover-crop may deprive the tree of nitrates when growth begins in the spring. In the long run, however, it may be a practical benefit on soils subject to considerable leaching by winter rains, because it takes up nitrates which would otherwise be leached away.

When the barley plants were about six to ten inches high, numerous tillers were produced by practically every plant. Many of the plants appeared to be making no growth during January. Early in February, however, it was observed that in practically every pot one tiller of each plant was gaining the ascendency, and making appreciable growth at the expense of the other tillers of the plant.

[1] STEWART, G. R., and MARTIN, J. C., Effect of various crops upon the water extract of a typical silty clay loam soil. Jour. Agric. Res. 20:663–667. 1921.

At the conclusion of the experiment each plant consisted of only one head-bearing culm, none of the other culms having survived. It is conceivable that plants of more than one culm and perhaps taller plants might have resulted, had fewer plants been grown in each pot. Twenty plants was the arbitrary number chosen, however, and may serve for the comparative requirements of the present experiment.

TABLE II

Plot	Fertilizer applied	Yield of barley			Average annual yield of citrus per tree in 3-year periods, 1918–1920 (in pounds)				
					Oranges		Lemons		Total
		Grain (gm.)	Straw (gm.)	Total dry wt. (gm.)	Navel	Valencia	Eureka	Lisbon	
B	Nothing	18.6	47.5	66.1	0	3	2	2	7
R	Potassium sulphate	21.1	40.3	61.4	0	3	3	2	8
M	Nothing	23.8	49.3	73.1	0	3	2	2	7
I	Potassium chloride	26.1	60.1	86.2	0	3	3	2	8
N	Superphosphate (blood)	27.5	69.5	97.0	.....	.....	.....	.....	.....
L	Nitrate, blood, potassium sulphate	29.6	91.0	120.6	28	19	4	26	77
P	Bone	30.2	76.9	107.1	1	21	3	31	56
D	Potassium sulphate	31.1	52.1	83.2	0	3	3	2	8
T	Nothing	39.3	75.0	114.3	0	3	2	2	7
E	Bone	43.1	118.0	161.1	1	21	3	31	56
K	Bone, potassium sulphate	43.5	82.9	126.4	1	21	3	31	56
J	Superphosphate	44.2	77.9	122.1	0	10	7	2	19
A	Complete (nitrate, blood, bone, potassium sulphate)	50.7	95.1	145.8	42	20	9	26	97
Q	Complete (nitrate, blood, bone, potassium sulphate)	74.2	155.3	229.5	42	20	9	26	97
Field 2, Box Springs	Nothing	74.3	146.3	220.6	.....	.....	.....	.....	.....
H	Nitrate of soda	78.8*	195.2	274.0	9	1	0	1	11
C	Blood	81.0	172.0	253.0	20	56	23	68	167
F	Manure	83.7	165.7	249.4	26	85	33	115	259
G	Nitrate, blood, bone	88.7	186.6	275.3	28	19	4	26	77
S	Blood	106.1	232.7	338.8	20	56	23	68	167
O	Manure, raw rock phosphate	129.7	216.3	346.0	26	85	33	115	259

* Grain conspicuously plump.

When the barley was harvested, the heads were first removed and beaten in a cloth bag. The grain was carefully cleaned and weighed. The plants were cut off close to the soil, and the entire tops, exclusive of grain, were weighed as straw. The results shown in table II represent the total air-dry weight for the three pots from each plot. For purposes of comparison with the growth of barley, the yields for citrus trees upon the various plots, together with their fertilizer treatment, are also given in table II (see footnote 2). This table shows the yield of grain to range from 18.6 to 129.7 gm.

Plots A, Q, H, C, F, G, S, and O had previously received nitrogenous fertilizers in larger amount (table III) than plots E, K, N, and P, and their soils produced a larger yield of grain. Plot L had received approximately the same amount of nitrogen as plots A, Q, etc., just mentioned, although its soil gave a yield of grain smaller than

TABLE III

ANNUAL FERTILIZER TREATMENT OF THE EXPERIMENTAL PLOTS

PLOT	FERTILIZER APPLIED	APPROXIMATE NUMBER OF POUNDS APPLIED PER ACRE PER ANNUM		
		N	P_2O_5	K_2O
A	Complete (nitrate,* blood, bone, potassium sulphate)	102	208	102
B	Nothing	.....	.....	.....
C	Blood	102	.....	.....
D	Potassium sulphate	.....	.....	102
E	Bone	34	208	.....
F	Manure (rough estimation)	102	76	140
G	Nitrate,* blood, bone	102	208	.....
H	Nitrate*	102	.....	.....
I	Potassium chloride	.....	.....	102
J	Superphosphate	.....	208	.....
K	Bone, potassium sulphate	34	208	102
L	Nitrate,* blood, potassium sulphate	102	.....	102
M	Nothing	.....	.....	.....
N	Superphosphate, blood	34	208	.....
O	Manure, raw rock phosphate (rough estimation)	102	208	140
P	Bone	34	208	.....
Q	Complete (nitrate,* blood, superphosphate, potassium sulphate)	102	208	102
R	Potassium sulphate	.....	.....	102
S	Blood	102	.....	.....
T	Nothing	.....	.....	.....
Field 2, Box Springs	Nothing†	.....	.....	.....

* Nitrate of soda.

† Twenty-five years continuous cropping to barley except two years to beans.

that obtained with soil from plots E and K, and about equal to that obtained with soil from plots N and P. Soil taken from plots D, T, and J, which received no nitrogenous fertilizer, produced a greater yield of grain than soil taken from some of the plots (N, L, P) which received nitrogen. In view of these facts we cannot conclude that the nitrogen added to these soils alone determines the

yield of barley grain which such soils are capable of producing. Other factors, at present not clearly defined, no doubt enter in and

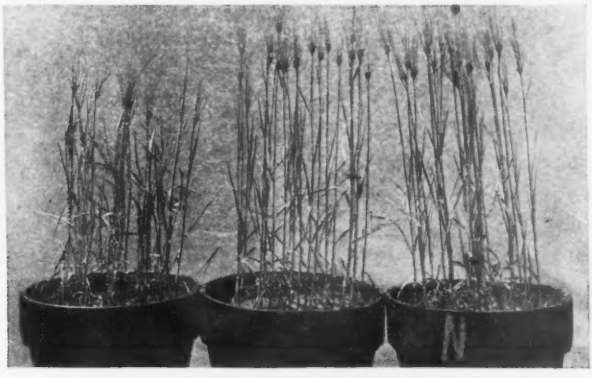

FIG. 1.—Growth of barley in soil from plot N

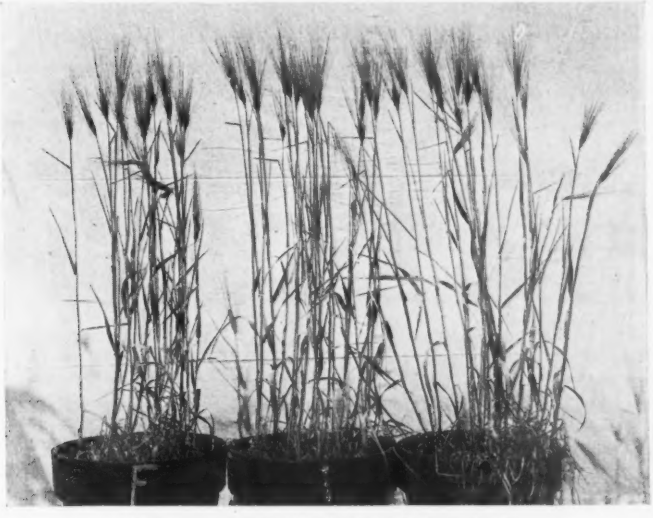

FIG. 2.—Growth of barley in soil from plot F

affect the processes of growth. The data seem to indicate, however, that the addition of potash or phosphoric acid to the plots has not

increased the capacity of their soils to produce barley. The same may be said regarding the effect of the addition of these two fertilizers to the plots upon the growth or fruiting conditions of the citrus trees.

If we compare the relative yields of grain with the fruit production of the trees on these plots, we find that plot L produced much less grain than would be assumed from its yield of citrus, were the

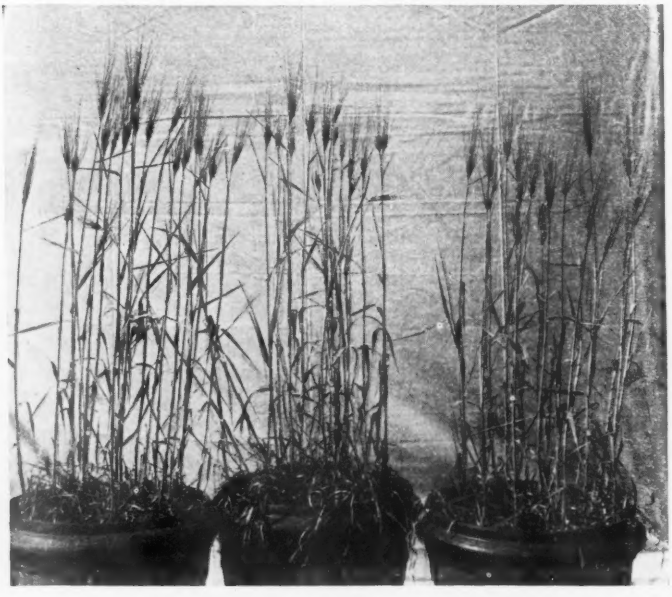

FIG. 3.—Growth of barley in soil from field no. 2, Box Springs tract; barley has been grown continuously on this soil for twenty-five years, except for two years in beans.

factors governing the growth of grain and citrus fruits considered to be the same. Manure, when applied to plots F and O, produced a pronounced beneficial effect upon the yield of citrus, and also greatly reduced the tendency toward mottle-leaf. The soil from plot F, however, produced less barley than soil from plots G and S, which received nitrogen from sources other than manure. Although the yield of barley on soil taken from plot H was quite

satisfactory and the grain conspicuously plump, the yields and the conditions of the citrus trees are very disappointing from an economic standpoint. The citrus trees on plot H were among the best trees in the experiment for the first seven years, but in the last seven years they have steadily deteriorated. The condition known as mottle-leaf developed to some extent before 1914, and has steadily become worse. At the present time the trees are in an extremely unhealthy condition, although, as indicated, the soil is capable of producing excellent barley. Nitrogen when added to

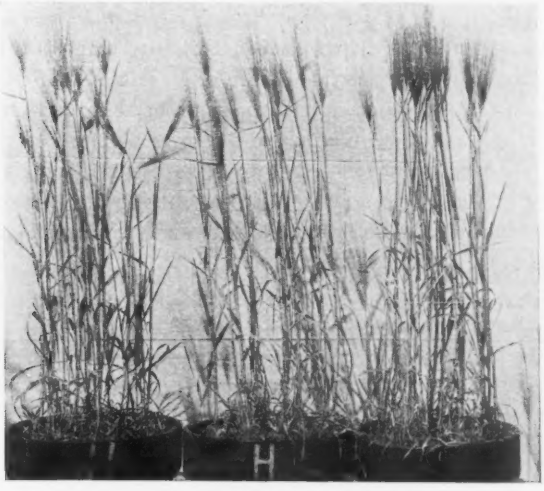

FIG. 4.—Growth of barley in soil from plot H

the plots in the form of sodium nitrate or dried blood, without additions of manure (plots A, C, G, H, L, Q, S), appears to intensify the mottling of the citrus trees, which ultimately is reflected in reduced yields from such trees.

Figs. 1–4 serve to illustrate the growth of barley produced on several of the soils. Fig. 1 shows the growth made in soil taken from plot N. The plants were short and rigid. The lower leaves had a tendency to die as material was transported to the upper parts of the plants. The heads were short and contained few seeds, which were hard and well-filled. Fig. 2 represents the growth

made upon soil obtained from plot F. Here the plants attained a good height and produced a satisfactory yield of grain. Fig. 3 shows the plants produced in soil from field no. 2, Box Springs tract, on which soil barley has been grown continuously for twenty-five years, except two years in beans. The plants produced on soil from plot H (Fig. 4) were quite similar to those shown in Fig. 3. Fig. 1 is fairly typical of the growth made in soils from plots B to A inclusive (table II); while Fig. 2 is quite representative of the growth made on soils from plots Q to O inclusive (table II).

Summary

The data presented in this paper show the lack of agreement between the barley producing power of soils and the yield and condition of citrus on such soils at the time the samples were taken. Moreover, it cannot be concluded that the amount of nitrogen previously added to a soil is an accurate measure of its barley producing power. The addition of phosphoric acid or potash to the soils studied has not shown any added advantages for the growth of barley or of citrus.

CITRUS EXPERIMENT STATION
RIVERSIDE, CAL.

CURRENT LITERATURE

BOOK REVIEWS

Fundamentals of fruit production

A textbook of more than usual merit has been prepared for students of horticulture by GARDNER, BRADFORD, and HOOKER.[1] The authors have given more attention to the fundamental physiological conditions necessary for successful growth and fruiting of trees, than to the art of planting and caring for orchards. The material is presented in seven sections, as follows: water relations, nutrition, temperature relations of fruit plants, pruning, fruit setting, propagation, and geographic influences in fruit production. The section on nutrition is particularly commendable.

The discussion is carried forward frequently by quotations from bulletins and periodic literature. Perhaps the authors have used this method somewhat too freely at times, for the student is likely to lose sight of the general significance of results in following the large number of quotations. One excellent feature of the work is the large number of literature citations for collateral readings at the close of each section. Nearly a thousand citations are thus listed, and intimate contact with the great mass of horticultural literature is made possible.

The book is a challenge to teachers of horticulture. Few students of this subject in the past have acquired the necessary fundamental training in plant physiology to make the best use of such a text. If its publication leads to the requirement of larger training for students of horticulture in fundamental physics, chemistry, and physiology, a great service to horticultural science has been rendered.—C. A. SHULL.

MINOR NOTICES

An Indian textbook of botany.—RANGACHARI[2] has published a revised and enlarged second edition of his text for Indian colleges. It differs chiefly from the first edition in added chapters on cryptogams, organic evolution, and heredity. It contains an unusual amount of technical material for an elementary college text, dealing chiefly with the morphology, physiology, and ecology of seed plants, the concluding chapters presenting briefly the lower groups in descending scale, ending with the fungi. One chapter on organic evolution and another on heredity and Mendelism conclude the text.—J. M. C.

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[1] GARDNER, V. R., BRADFORD, F. C., and HOOKER, H. D., The fundamentals of fruit production. 8vo. pp. xvi+686. New York: McGraw-Hill. 1922.

[2] RANGACHARI, RAI BAHADUR K., A manual of elementary botany for India. 2d ed. 8vo. pp. xx+572. *figs. 494.* 1921.

NOTES FOR STUDENTS

Taxonomic notes.—BRAINERD and PEITERSEN[3] have investigated the polymorphic genus *Rubus* as represented in New England, especially Vermont, to determine the origin of the numerous polymorphic forms and their relationship to one another. They have not only examined all available herbarium material, but also have studied the plants intensively in the field and in the experimental garden. The results are full of interest, showing the result of environment and of hybridization in the production of species. An interesting conclusion is stated as follows: "It seems to us that it is no longer a question of whether or not new species of blackberries are produced in the wild through natural hybridization, but rather that it is a question whether hybridization is not the primary and only factor in the production of new species within this group." The taxonomic statement of the results are given for the twelve recognized New England species of the section EUBATUS, all of which are illustrated by plates. There is no genus about which taxonomists have been more discordant in their opinions, which has been due, in the opinion of the authors, to "too great reliance upon herbarium specimens and a failure to study sufficiently the life history of growing plants in field and in garden cultures, failure to appreciate the variation in the supposed specific characters of these plants due entirely to the environmental conditions, and lack of appreciation of the extent of interbreeding of the blackberries in the wild, and imperfect knowledge of the laws of inheritance, in accordance with which opposed parental characters are transmitted to their hybrid offspring."

MAXON,[4] in continuation of his studies of tropical American ferns, has described 13 new species and a new genus (*Atalopteris*) related to *Dryopteris*.

BLAKE[5] has published a revision of the South American genus *Diplostephium* (related to *Aster*), recognizing 41 species, 13 of which are described as new.

FAIRMAN[6] has described 37 new species of fungi, 22 of which belong to the Deuteromycetes, and 10 to the Ascomycetes. *Gamonaemella* is described as a new genus of Deuteromycetes, and *Amblyosporiopsis* as a new genus related to *Amblyosporium*.

PETCH,[7] in his second contribution of additions to Ceylon fungi, has described 107 new species and the following new genera: *Dendrocyphella* (Thelephoraceae) and *Phaeoglossum* (Geoglossaceae).

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[3] BRAINERD, EZRA, and PEITERSEN, A. K., Blackberries of New England, their classification. Vermont Agric. Exper. Station Bull. 217. pp. 84. *pls. 36*. 1920.

[4] MAXON, W. R., Studies of tropical American ferns. no. 7. Contrib. U.S. Nat. Herb. 24:33–63. *pls. 11–20*. 1922.

[5] BLAKE, S. F., Key to the genus *Diplostephium*, with descriptions of new species. Contrib. U.S. Nat. Herbarium 24:65–86. *pls. 21–24*. 1922.

[6] FAIRMAN, C. E., New or rare fungi from various localities. Proc. Rochester Acad. Sci. 6:117–139. *pls. 21–23*. 1922.

[7] PETCH, T., Additions to Ceylon fungi (II). Ann. Roy. Bot. Gard. Peradeniya 7:279–322. 1922.

STANDLEY[8] has published a second instalment of the *Trees and shrubs of Mexico*, including 45 families, represented by 110 genera and 1487 species. The publication brings together and organizes much recent taxonomic work upon these groups, and also presents the economic uses of many species. Only 17 new species are described in almost as many genera, the purpose being chiefly to present additional valuable information concerning Mexican plants. The genus *Quercus* is presented by TRELEASE, who remarks that "no other country has so large a number of species," the list including 112. One of the large and very interesting genera is *Phoradendron*, with 57 species. The great display, however, is that of the Leguminosae, presented as three families, represented by 85 genera and 751 species. The largest genus is *Parosela* (106 spp.), and following it are *Mimosa* (67 spp.), *Acacia* (64 spp.), and *Cassia* (51 spp.).

BURT[9] has published a critical account of 29 North American species of Tremellaceae, Dacryomycetaceae, and Auriculariaceae, including 9 new combinations and 3 new species.

DE WILDEMAN[10], in continuation of his study of the African species of *Alchemilla*, has described 14 new species. The enumeration of species shows that the contributions from South Africa have made this one of the large genera.

CHARDON[11] has published an account of the Pyrenomycetes of Porto Rico, including 15 genera represented by 27 species, 9 of which are described as new. The interest of American mycologists in the mycological flora of this island and of the West Indies in general is resulting in a steady increase of our information.

In continuation of the systematic account of the plants collected in New Caledonia and the Isle of Pines by COMPTON in 1914, the following groups have been presented:[12] Hepaticae, by WM. H. PEARSON (28 new species in 19 genera); Freshwater algae, by NELLIE CARTER, including a new genus (*Rosaria*) of Stigonemaceae; Lichens, by A. LORRAIN SMITH, including 20 new species and a new genus (*Lepidoleptogium*) of Pannariaceae; Fungi, by ELSIE M. WAKEFIELD, including 33 species; and Mycetozoa, by G. LISTER, represented by 10 species.

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[8] STANDLEY, PAUL C., Trees and shrubs of Mexico (Fagaceae-Fabaceae). Contrib. U.S. Nat. Museum **23**:171–515. 1922.

[9] BURT, E. A., Some North American Tremellaceae, Dacryomycetaceae, and Auriculariaceae. Ann. Mo. Bot. Gard. **8**:361–396. *pls. 3.* 1921.

[10] DE WILDEMAN, E., Additions a la flore du Congo. Bull. Jard. Bot. Bruxelles **7**:271–386. 1921.

[11] CHARDON, C. E., A contribution to our knowledge of the Pyrenomycetes of Porto Rico. Mycologia **13**:279–300. *pls. 13–15.* 1921.

[12] A systematic account of the plants collected in New Caledonia and the Isle of Pines by Mr. R. H. COMPTON, M.A., in 1914. Part III. Cryptogams (Hepaticae-Fungi). Jour. Linn. Soc. **46**:13–96. *pls. 2–5.* 1922.

ARTHUR,[13] in connection with a description of new species of Uredineae, has described a new genus (*Diabole*), which had been assigned to *Uromycladium*. It occurs in Cuba on *Mimosa pigra*.

BURT[14] has published a detailed monograph of the North American species of *Clavaria*, recognizing 98 species, 4 of which are described as new. In addition, 16 species are listed as imperfectly known, 8 species are excluded, and 6 tropical species are described. It is interesting to note that of the 98 North American species described, 33 are credited to PECK and 19 to ATKINSON.

BRITTON and ROSE[15] have segregated two new genera (*Thelocactus* and *Neolloydia*) from *Echinocactus*.

TURRILL[16] has described a new genus (*Isotheca*) of Acanthaceae from Trinidad, related to *Herpetacanthus*.

ULBRICH[17] has described two new genera (*Rhopalopodium* and *Aspidophyllum*) of Ranunculaceae from South America.

ROWLEE[18] has monographed the genus *Costus* (Zingiberaceae) as represented in Central America. He recognizes 14 species, 3 of which are described as new. "As at present organized, the genus *Costus* is composed of about 100 species, divided almost equally between the eastern and western hemispheres."

THARP[19] has described a new genus (*Commelinantia*) of Commelinaceae from Texas. It had been referred to *Tradescantia* by TORREY, as *T. anomala*.

WINKLER,[20] in continuation of his monograph of the Urticaceae of New Guinea, has described 59 new species, 33 of which belong to *Elatostema*.

ENGLER,[21] with the cooperation of a group of taxonomists, in continuation of the investigation of the flora of Africa, has published 65 new species. *Freyliniopsis* is established as a new genus of Scrophulariaceae, and 32 of the new species belong to *Mesembrianthemum*.

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[13] ARTHUR, J. C., New species of Uredineae. XIV. Bull. Torr. Bot. Club **49**:189–196. 1922.

[14] BURT, E. A., The North American species of *Clavaria*, with illustrations of the type specimens. Ann. Mo. Bot. Gard. **9**:1–78. *pls. 1–11*. 1922.

[15] BRITTON, N. L., and ROSE, J. N., Two new genera of Cactaceae. Bull. Torr. Bot. Club **49**:251, 252. 1922.

[16] TURRILL, Decades Kewenses CVI. Kew Bull. no. 6. 1922.

[17] ULBRICH, E., Ranunculaceae novae vel criticae. V. Notizbl. Bot. Gart. Berlin-Dahlem **8**:251–272. 1922.

[18] ROWLEE, W. W., The genus *Costus* in Central America. Bull. Torr. Bot. Club **49**:283–292. *pls. 12–15*. 1922.

[19] THARP, B. C., *Commelinantia*, a new genus of the Commelinaceae. Bull. Torr. Bot. Club **49**:269–275. *pls. 10, 11*. 1922.

[20] WINKLER, H., Die Urticaceen Papuasiens. Bot. Jahrb. **75**:529–608. 1922.

[21] ENGLER, A., Beitrage zur Flora von Afrika. XLIX. Bot. Jahrb. **75**:609–648. 1922.

DAVY,[22] in a revision of the South African species of *Dianthus* (Caryophyllaceae), recognizes 17 species, 6 of which are described as new. He says that the genus, "as represented in South Africa, has long been a source of trouble to systematists."

THAXTER[23] has published a revision of the Endogoneae. He states that the fungi of this group are "rather infrequently met with, owing in part to their apparent rarity, and partly to the fact that certain of the species, at least, are truly hypogaeous, and may develop at a depth of several inches below the ground, or beneath thick mats of Sphagna or other mosses." In presenting the group, 4 genera are recognized, including 24 species. The large genus is *Endogone*, with 19 species, 9 of which are described as new.

THAXTER[24] has also published an account of two remarkable Ascomycetes. They are a new species of *Hypocreopsis* from North Carolina and East Tennessee, growing on stems and branches of a *Rhododendron*, and a new genus (*Medeolaria*) from New England, forming distortions on a species of *Medeola*.

SETCHELL and GARDNER,[25] in continuation of their investigation of the algae of the Pacific coast, have described new species in the following genera: *Myrionema* (8 spp.), *Compsonema* (15 spp.), *Hecatonema* (3 spp.), *Pylaiella* (2 spp.), *Streblonema* (11 spp.), and *Ectocarpus* (13 spp.).—J. M. C.

Cytology of hybrids in Papaver.—Interspecific hybrids provide one of the most promising problems of botany, and one whose investigation is of particular interest to the cytologist. The peculiarities of the reduction division, while explaining why gametes so often fail to function, may also throw some light on the possibility of polyploidy arising in this way, and may assist in clearing up obscure points in the normal process of meiosis. A preliminary report by LJUNGDAHL[26] of his work on *Papaver*, while listing twelve crosses made by him, six of which had not been made previously, deals exclusively with the reduction division in the pollen mother cells of F_1 hybrids of the two crosses, *P. atlanticum*×*P. dubium*, and *P. somniferum*×*P. orientale*. Reciprocal matings are not mentioned. The haploid numbers of chromosomes found in this genus are 7, 14, 21, 35, 11, and 22. With the exception of the last two, the relation of these numbers appears significant, and suggests the production of all

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[22] DAVY, J. BURTT, A revision of the South African species of *Dianthus*. Kew Bull. no. 7. 1922.

[23] THAXTER, ROLAND, A revision of the Endogoneae. Proc. Amer. Acad. **57**:291–350. *pls. 4*. 1922.

[24] ———, Note on two remarkable Ascomycetes. Proc. Amer. Acad. **57**:425–436. *pls. 2*. 1922.

[25] SETCHELL, W. A., and GARDNER, N. L., Phycological Contributions II to VI. Univ. of Calif. Publ. Bot. **7**:333–426. *pls. 32–49*. 1922.

[26] LJUNGDAHL, HILDUR, Zur Zytologie der Gattung *Papaver*. Svensk Botanisk Tidskrift **16**:103–114. *fig. 6*. 1922.

these species from one source through polyploidy. Hybridization would appear a promising way of testing this hypothesis.

In the first cross the haploid chromosome numbers are 7 and 14 respectively. There appeared to be little affinity between the chromosome groups, and great irregularities in the reduction division were apparent. The hybrid is completely sterile, degeneration being noted at all stages of spore formation. Moreover, no uniform rate of division was observed. Sometimes in the heterotypic mitosis no spindle is formed, there being a direct transition from diakinesis to interkinesis. This large unreduced nucleus can then undergo the homotypic division with probable equal apportionment of chromosomes to the daughter nuclei. More frequently, however, a spindle is formed and 5–7 chromosomes go to the poles, leaving the remainder in the equatorial region to be later distributed more or less equally to both poles. An irregular distribution has also been observed, often resulting in two daughter nuclei united by a bridge. The homotypic division also shows all kinds of irregularities. Sometimes "delayed" chromosomes are observed in the equatorial region during anaphase, and often, as has been noted in other hybrids and in apogamous plants, a common nuclear membrane surrounds the whole spindle, forming one nucleus, or perhaps two, united by a bridge.

The chromosomes of the hybrid *P. somniferum*×*P. orientale*, 11 from the female parent and 21 from the male, show greater affinity than in the preceding case. In the heterotypic division there can usually be counted 7–9 distinct paired chromosomes in diakinesis, and sometimes 11 can be distinguished. The remaining univalents lie scattered irregularly when the spindle is formed, they undergo a longitudinal split, and usually the split halves are distributed to the poles along with the separated chromosomes of the pairs. Each daughter nucleus is thus made up of 11 "whole" and 10 "half" chromosomes. Irregularities of distribution have been observed, however, both as to the "whole" and the "half" chromosomes, together with other unusual conditions.

In the homotypic mitosis the 10 chromosomes are "delayed," as was found in the previous cross, but apparently 5 go to each pole in most cases. Sometimes both homotypic spindles are joined side by side, all stages between "double" and "single" spindles being found, and belated chromosomes being in evidence as before. This presumes an irregular heterotypic division, and telophases were found in which the daughter nuclei were united by a chromosome strand. The double spindle homotypic division results in cells, each with 32 chromosomes, and if they were functional they would produce diploid gametes. Such diploid cells can also arise by the two spindles being in contact at one pole, thus producing three nuclei, one of which is diploid. Such cases were frequently found.

While the gametes of this hybrid are mostly non-functional, still it appears to the writer, since this is a method of diploid gamete formation without apparent great disturbance of the nuclear division mechanism, that appropriate plants might thus give rise to polyploid forms.—R. O. EARL.

Permeability.—COLLANDER[27] has investigated the permeability of plant cells to a number of sulphonic acid dyes. The method consisted in placing sections of the tissue in concentrated solutions of the dyes for a period varying from one to several days. The sections then were mounted on a slide in solutions of the dye of varying concentrations, and it was determined by observing through the microscope in which concentration the interior of the cell was as darkly colored as the surrounding solution. In this way it was found that, in the case of most of the dyes worked with, the concentration of the dye in the cells was from 8 to 160 times smaller than the concentration of the dye in the surrounding solution. By appropriate experiments it was shown that this was really a case of the dyes not entering the cells. The possibility that the dyes entered the cells quite readily, and then were decolorized by reduction or other chemical process, or had been adsorbed by the cell colloids, was shown to be not true. It was also shown that the establishment of equilibrium conditions between the concentration of the dye on the inside and that on the outside of the cells, with the latter concentration much greater than the former, is due to the impermeability of the cells or protoplasts to the dye, and not to certain physical-chemical equilibria, which sometimes occur.

Certain exceptions to the rule of the impermeability of plant cells to the dyes were observed. Some of these were flower petals, young embryo cells, and cells immediately next to the vascular bundles. These were found to be quite permeable to the dyes. In most of the tissue worked with there were certain cells, sometimes quite abundant, which the dyes entered readily. Although these cells appeared sound, on close examination it was usually found that they were cells of impaired vitality. It is believed that the presence of these cells in plant tissue has led certain investigators to conclude that sulphonic acid dyes enter plant cells readily. Low temperatures, narcotics, and hydroxyl ions were found to lessen the entrance of the dyes. This was not true, however, of trivalent cations. The data in regard to the taking in of sulphonic acid dyes are discussed in reference to the various permeability theories, and are shown to be more in accord with OVERTON's lipoid theory than with any of the other theories.

The experimental work described in this paper seems to have been very carefully done, and the conclusions arrived at only after the exclusion of all other possibilities.—S. V. EATON.

Germination and polyembryony of Poa.—NISHIMURA[28] has published a preliminary note of the results of his investigations of the comparative morphology and development of *Poa pratensis*, *Phleum pratense*, and *Setalia italica*.

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[27] COLLANDER, RUNAR, Über die Permeabilität pflanzlicher Protoplasten für Sulfosäurestoffe. Jahrb. Wiss. Bot. **60**:354–410. 1921.

[28] NISHIMURA, MAKOTO, On the germination and the polyembryony of *Poa pratensis* L. Bot. Mag. Tokyo **36**:47–54. *pl. 1.* 1922.

The present report deals with the germination and polyembryony of *Poa pratensis*. One of the interesting features in the germination of this species is the development of numerous hairs on the coleorhiza, corresponding in origin and structure to ordinary root hairs. It is concluded that these coleorhiza hairs "serve as a unique device for putting the embryo in immediate contact with the soil water, and so tide it over the period required for the development of an absorbing root surface." It is also concluded that the epiblast, with its associated structure, is a cotyledon, a confirmation of the recently developed views as to the origin of monocotyledony. The occurrence of polyembryony was studied, various types being noted, but the most common expression of it was the development of two or three embryos from a common massive suspensor. In some instances the egg did not function at all, and all the embryos arose from other sources. The conclusion is that polyembryony and other irregularities are usually associated with ovaries that have become abnormal as a result of the sting of insects which deposit their eggs in the ovary at various stages of its development. Just what the nature of the resulting stimulus is was not determined, but the possibility of changes of osmotic pressure is suggested.—J. M. C.

Nitrogen fertilizers.—KUYPER[29] has given the results of the use of nitrogen fertilizers for sugarcane cultivation in Java. Under the supervision of the Sugar Experiment Station at Pasoeroean, field trials have been made during many years on a great number of sugar plantations as to the relative value of nitrogen fertilizers. Sulphate of ammonia, being the generally used fertilizer (the average amount is about 380 lb. per acre), is used as the basis for comparison. During the last ten years, 162 comparisons were made between sulphate of ammonia and nitrate of soda. These two fertilizers proved to be of the same value, the sulphate of ammonia being preferable, because it is not hygroscopical. Nitrolim or cyanamid is not so good as sulphate of ammonia. In about 100 field trials the latter manure gave in twenty-four of the tests the same result as nitrolim, and in fifty-three of the tests a better one. "Ammonsulfaatsalpeter," imported by the "Badische Anilin und Soda fabriken," proved to be too hygroscopical for the condition in the tropical rainy season. "Ureum," also imported by the "Badische," was compared with sulphate of ammonia in twelve field trials; the results were the same. It is expected to be a good substitute for $(NH_4)_2SO_4$. A more detailed study is made on beancake, the residue by oil extraction from *Arachis hypogaea*. This fertilizer is not so good as sulphate of ammonia, being better on light soils than on sandy soils. A combination of beancake and sulphate of ammonia is better than the application of beancake only. In all the experiments the same weight of nitrogen was given in the different manures.—J. KUYPER.

[29] KUYPER, J., De waarde van de verschillende stikstofmeststoffen voor de suikercultuur op Java (The relative value of several nitrogen fertilizers for sugarcane cultivation in Java). Arch. Suikerindustrie in Nederlandsch-Indie. 1922. Mededeelingen no. 3. pp. 145–154. 1922.

Distribution of sex in angiosperms.—C. and H. YAMPOLSKY[30] have published a very detailed and tabulated account of the distribution of sex forms among the angiosperms. The data are organized under 43 classes of sex combinations. It appears that about 70 per cent of angiosperms are listed as strictly hermaphrodite. In approximately one-fourth of the genera, therefore, manifold sex combinations occur. The polygamous forms are more general among monocotyledons than dicotyledons, appearing in 73 and 42 per cent of the orders respectively. The charts show, however, that the variety of combinations of different sex forms is much greater in dicotyledons than in monocotyledons. Very interesting results are obtained from noting the relative distribution of the groups of sex forms in the evolutionary scheme. It was discovered, for example, that the larger percentage of hermaphrodites are distributed among the orders nearer the end of the evolutionary series. Many interesting conclusions are drawn from the comparison of orders in this particular.

The whole contribution is an organized assemblage of useful and suggestive data, which a brief notice cannot include, but which should be available to students of evolution and genetics.—J. M. C.

Pollination of Marcgravia.—BAILEY[31] has been examining species of the orchid genus *Marcgravia* in British Guiana, and has challenged the old notion of its adaption to ornithophily. He says "the bizarre inflorescenes of *Marcgravia*, with their curious pitcher-like nectaries, are assumed to facilitate cross-pollination by birds. In fact, this extraordinary genus is cited as one of the most striking illustrations of ornithophily." His observations indicate that there is no reliable evidence that birds are concerned in the pollination of the flowers. The highly specialized inflorescences are not efficient mechanisms for insuring pollination by humming birds, the parts being so arranged that visiting birds do not become coated with pollen. The flowers of the two species studied in detail, which are described as new species (*M. cuyuniensis* and *M. purpurea*), appear to be self-pollinated. The physiological significance, therefore, of this elaborate floral mechanism remains to be discovered.—J. M. C.

Eccentric growth and redwood formation in conifers.—BURNS[32] has investigated experimentally the factors causing the production of eccentric growth and the formation of redwood in conifers. These phenomena have been explained as due to various causes, as compression by wind, differences in illumination, differences in branching, and gravity acting as a stimulus. These conclusions

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[30] YAMPOLSKY, CECIL and HELENE, Distribution of sex forms in the phanerogamic flora. Bibl. Genetica 3:1–62. charts 8. 1922.

[31] BAILEY, I. W., The pollination of *Marcgravia:* a classical case of ornithophily? Amer. Jour. Bot. 9:370–384. *pls. 23, 24. figs. 5.* 1922.

[32] BURNS, G. P., Eccentric growth and the formation of redwood in the main stem of conifers. Vermont Agric. Exper. Sta. Bull. 219. pp. 16. *figs. 10.* 1920.

are based on many field observations and very little experimental work. As a result of his experiments, the author concludes that redwood and eccentric growth usually but not necessarily occur together; that compression does not act as a stimulus causing the cambium to divide more rapidly and produce redwood; that the production of redwood in the trees examined is a "morphogenic response" to gravitation stimulus.—J. M. C.

Morphology of Martynia.—Miss ANDERSON[33] has investigated the floral development and embryogeny of *Martynia louisiana*. The embryo sac is long and narrow, the large egg extending far down into the sac cavity. The three antipodals are long slender cells that increase in size and are persistent. The period between pollination and fertilization is unusally short, being only six to eight hours. The first division of the endosperm cell occurs before fertilization, and the bulk of the endosperm is in the lower part of the sac where the embryo develops. The suspensor is very long, and pushes the embryo into the lower portion of the sac. The development of the embryo is said to be like that of *Capsella*.—J. M. C.

Endosperm of cereals.—Miss GORDON[34] has investigated the development of endosperm in the common cereals, and has reached some interesting conclusions. The early layer of endosperm lining the embryo sac assumes the character of a cambium, producing cells only on its inner surface. After these cambium cells have ceased to divide, they become filled with aleurone grains, forming the aleurone layer. The greater respiratory activity of this layer and the presence of vitamines in it are the natural results of its being a resting cambium. "Whether it can be awakened to further activity during germination remains for future investigation."—J. M. C.

An unusual Spirogyra.—Miss MERRIMAN[35] has described a *Spirogyra*, collected in one of the parks of New York City, in which the chloroplasts become parallel in conjugating filaments. In non-conjugating filaments they are more or less spirally arranged. It is regarded as a new species, and named *S. rectispira*, ranking "with *S. crassa* in the diameter of its filaments as the largest of the genus."—J. M. C.

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[33] ANDERSON, FLORA, The development of the flower and embryogeny of *Martynia louisiana*. Bull. Torr. Bot. Club **49**:141–157. *pls. 7, 8. figs. 43.* 1922.

[34] GORDON, MARY, The development of endosperm in cereals. Proc. Roy. Soc. Victoria **34**:105–116. *figs. 9.* 1922.

[35] MERRIMAN, MABEL L., A new species of *Spirogyra* with unusual arrangement of the chromatophores. Amer. Jour. Bot. **9**:283, 284. *figs. 3.* 1922.

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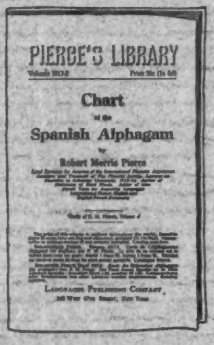

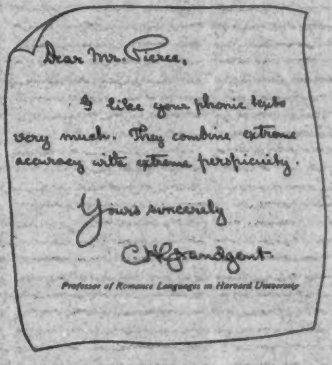
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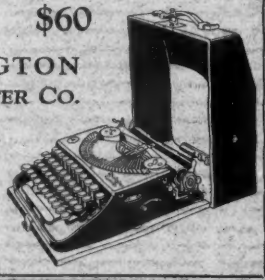

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